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(54) METHOD FOR PRODUCING L-AMINO ACID

VERFAHREN ZUR HERSTELLUNG VON L-AMINOSÄURE

PROCEDE DE PRODUCTION D ACIDE L-AMINE

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Description

Technical Field

[0001] The present invention relates to a method for efficiently producing L-amino acids such as L-glutamic acid, L-glutamine, L-proline and L-arginine, by fermentation.

Background Art

[0002] L-amino acids have been produced mainly by various fermentation methods using L-amino acid-producing coryneform bacteria, including those belonging to the genus *Brevibacterium, Corynebacterium, Microbacterium,* or mutant strains thereof (Amino Acid Fermentation, Japan Scientific Societies Press, p.195-215,1986). Examples of known methods for producing an L-amino acid by fermentation using other bacteria include a method using a microorganism belonging to the genus *Bacillus, Streptomyces,* or *Penicillium* (US Patent 3,220,929), a method using a bacterium belonging to the genus *Pseudomonas, Arthrobacter, Serratia,* or *Candida* (US Patent 3,563,857), a method using a bacterium belonging to the genus *Bacillus, Pseudomonas, Serratia,* or *Aerobacter aerogenes* (currently, *Enterobacter aerogenes*) (JP 32-9393 B), and a method using a mutant strain of *Escherichia coli* (US Patent 5378616). Furthermore, methods for producing an L-amino acid using bacteria belonging to the genus *Klebsiella, Erwinia, Pantotea,* or *Enterobacter* (EP 955368 A, EP 952221 B, and EP 999282 A) have also been disclosed.

[0003] Various methods to increase L-amino acid-producing ability by enhancing activities of L-amino acid biosynthetic enzymes by recombinant DNA techniques have also been disclosed. For example, it has been reported that introducing a gene encoding citrate synthase derived from *Escherichia coli* or *Corynebacterium glutamicum* into a bacterium belonging to the genus *Corynebacterium* or *Brevibacterium* is effective to enhance the L-amino acid-producing ability of the bacterium (JP 07-121228 B). In addition, it has been reported that introducing a citrate synthase gene derived from a coryneform bacterium into an enterobacterium belonging to the genus *Enterobacter, Klebsiella, Serratia, Erwinia,* or *Escherichia* is effective to enhance L-glutamic acid-producing ability (EP 999282 A).

[0004] Phosphotransacetylase is an enzyme which is involved in acetic acid metabolism. In *Escherichia coli,* this enzyme plays a role in the reaction which produces acetyl phosphate from acetyl-CoA and phosphate, which is part of the primary pathway for producing acetic acid. Furthermore, it is known that in *Corynebacterium glutamicum,* phosphotransacetylase activity increases upon production of acetyl-CoA by assimilating acetic acid, and the activity is negatively regulated by the transcription factor RamB (Microbiology 1999 503-513, Journal of Bacteriology 2004 vol.186, No.9 p2798-2809).

Examples of known methods of fermentative production of a useful material using a bacterium in which phosphotransacetylase activity is enhanced include production of poly-β-hydroxyalkanoate copolymer (US Patent 5,891,686 and US Patent 5,569,595), and production of ethanol (WO 2003/078643). Furthermore, an enzymatic method of producing a sulfur-containing L-amino acid using phosphotransacetylase has been disclosed (JP 09-009982 A). However, enhancing phosphotransacetylase activity in the breeding of L-amino acid-producing bacteria has not been reported, and the relationship between the phosphotransacetylase activity and L-amino acid productivity has not been elucidated.

DISCLOSURE OF THE INVENTION

[0005] An object of the present invention is to provide a novel fermentation method for producing L-amino acids such as L-glutamic acid, L-glutamine, L-proline and L-arginine.

[0006] The inventors of the present invention have made extensive studies to achieve the above-described object. As a result, they have found that L-amino acids such as L-glutamic acid, L-glutamine, L-proline and L-arginine can be efficiently produced by culturing a L-amino acid-producing bacterium which has been modified to enhance phosphotransacetylase activity, and thereby completed the present invention.

[0007] It is an object of the present invention to provide a method for producing an L-amino acid comprising culturing a L-amino acid-producing bacterium which has been modified to have enhanced phosphotransacetylase activity in a medium, and collecting the L-amino acid from the medium or the bacterium.

[0008] It is another object of the present invention to provide the method as described above, wherein the phosphotransacetylase activity is enhanced by increasing the copy number of the gene encoding phosphotransacetylase or by modifying an expression regulatory sequence of the gene encoding phosphotransacetylase.

[0009] It is another object of the present invention to provide the method as described above, wherein the gene encoding phosphotransacetylase is a DNA selected from the group consisting of:

(a) a DNA comprising nucleotides 1214 to 2641 of SEQ ID NO: 34 or the nucleotide sequence of SEQ ID NO: 40; and
(b) a DNA which hybridizes with a nucleotide sequence which is complementary to the nucleotide sequence of

nucleotides 1214 to 2641 of SEQ ID NO: 34 or the nucleotide sequence of SEQ ID NO: 40, wherein said DNA hybridizes under stringent conditions and encodes a protein that has phosphotransacetylase activity.

[0010] It is another object of the present invention to provide the method as described above, wherein the phosphotransacetylase activity is enhanced by disrupting a ramB gene.

[0011] It is another object of the present invention to provide the method as described above, wherein the bacterium is further modified so that an activity is enhanced of a protein selected from the group consisting of D-xylose 5-phosphate-phosphoketolase, fructose 6-phosphate phosphoketolase, and combinations thereof.

[0012] It is another object of the present invention to provide the method as described above, wherein the bacterium is further modified to have enhanced pyruvate carboxylase activity.

[0013] It is another object of the present invention to provide the method as described above, wherein the bacterium is further modified to have enhanced phosphoenolpyruvate carboxylase activity.

[0014] It is another object of the present invention to provide the method as described above, wherein the bacterium is selected from the group consisting of a coryneform bacterium, *Pantoea* bacterium, *Enterobacter* bacterium, and *Escherichia* bacterium.

[0015] It is another object of the present invention to provide the method as described above, wherein the L-amino acid is selected from the group consisting of L-glutamic acid, L-glutamine, L-proline and L-arginine.

BRIEF DESCRIPTION OF THE DRAWINGS

[0016] Fig. 1 shows an IS-inserted site in the mutant-type yggB gene.

DESCRIPTION OF THE PREFERRED EMBODIMENTS

<1-1> Bacterium of the present invention

[0017] The bacterium used in the production method of the present invention (also referred to as the bacterium of the present invention) has an L-amino acid-producing ability and has been modified to have enhanced phosphotransacetylase activity. The bacterium of the present invention can be obtained by modifying a bacterium having an L-amino acid-producing ability (i.e., parental strain) so that the phosphotransacetylase activity is enhanced. The bacterium of the present invention may have a native L-amino acid-producing ability or may be bred to have an L-amino acid-producing ability.

[0018] Herein, the "L-amino acid-producing ability" means an ability to produce an L-amino acid in an amount that allows for collection of the L-amino acid from the bacterial cells or the medium. Preferably, it means an ability to produce a larger amount of the L-amino acid as compared to a wild-type or non-modified strain which is cultured under the same conditions.

[0019] Examples of the L-amino acids to be produced include, L-glutamic acid, L-glutamine, L-arginine and L-proline.

[0020] Examples of the bacterium of the present invention include, but are not limited to, bacteria belonging to the *Enterobacteriaceae* family, including those belonging to the genus *Escherichia, Pantoea, Enterobacter,* or the like, coryneform bacteria such as *Corynebacterium glutamicum* and *Brevibacterium lactofermentum,* and bacteria belonging to the genus *Bacillus,* such as *Bacillus subtilis.*

[0021] In the present invention, the "coryneform bacterium" includes bacteria previously classified as *Brevibacterium,* but now are classified as *Corynebacterium* (Int. J. Syst. Bacteriol., 41, 255 (1991)), as well as bacteria belonging to the genus *Brevibacterium,* which are highly-related to the genus *Corynebacterium.* Examples of such bacteria include the following:

[0022]

Corynebacterium acetoacidophilum
Corynebacterium acetoglutamicum
Corynebacterium alkanolyticum
Corynebacterium callunae
Corynebacterium glutamicum
Corynebacterium /ilium
Corynebacterium melassecola
Corynebacterium thermoaminogenes (Corynebacterium efficiens)
Corynebacterium heraculis
Brevibacterium divaricatum
Brevibacterium flavum

Brevibacterium immariophilum
Brevibacterium lactofermentum (Corynebacterium glutamicum)
Brevibacterium roseum
Brevibacterium saccharolyticum
Brevibacterium thiogenitalis
Corynebacterium ammoniagenes
Brevibacterium album
Brevibacterium selinum
Microbacterium ammoniaphilum

[0023] Specifically, the following strains are exemplified:

Corynebacterium acetoacidophilum ATCC13870
Corynebacterium acetoglutamicum ATCC15806
Corynebacterium alkanolyticum ATCC21511
Corynebacterium callunae ATCC 15991
Corynebacterium glutamicum ATCC13020, ATCC13032, ATCC13060
Corynebacterium lilium ATCC15990
Corynebacteruim melassecola ATCC 17965
Corynebacterium thermoaminogenes AJ12340 (FERM BP-1539)
Corynebacterium herculisATCC13868
Brevibacterium divaricatum ATCC 14020
Brevibacterium flavum ATCC 13 826, ATCC 14067
Brevibacterium immariophilum ATCC14068
Brevibacterium lactofermentum ATCC13869 *(Corynebacterium glutamicum* ATCC13869)
Brevibacterium roseum ATCC13825
Brevibacterium saccharolyticum ATCC14066
Brevibacterium thiogenitalis ATCC19240
Corynebacterium ammoniagenes ATCC6871, ATCC6872
Brevibacterium album ATCC15111
Brevibacterium selinum ATCC15112
Microbacterium ammoniaphilum ATCC15354

[0024] These strains are available from the American Type Culture Collection (ATCC: address: P.O. Box 1549, Manassas, VA 20108, 1, United States of America). That is, accession numbers are given to each of the strains, and the strains can be ordered using these numbers (http://wwwatcc-.org/). The accession numbers for the strains are listed in the catalogue of the American Type Culture Collection. The AJ12340 strain was deposited at the National Institute of Bioscience and Human-Technology, Agency of Industrial Science and Technology (currently, International Patent Organism Depositary, National Institute of Advanced Industrial Science and Technology) (Central 6, 1-1-1 Higashi, Tsukuba, Ibaraki 305-5466, Japan) on October 27, 1987 and given an accession number of FERM BP-1539 under the provisions of Budapest Treaty.

[0025] Bacteria belonging to the *Enterobacteriaceae* family are not particularly limited as long as they have an L-amino acid-producing ability, and include those belonging to the genus *Escherichia, Enterobacter, Pantoea, Klebsiella, Serratia, Erwinia, Salmonella, Morganella,* or the like. Bacteria belonging to the *Enterobacteriaceae* family based on the classification described in the NCBI (National Center for Biotechnology Information) database (http://www.ncbi.nlm.nih.gov/htbin-post/Taxonomy/wgetorg?mode=Tree&id=1236&lvl=3& keep=1 &srchmode= &unlock) can be used. Among these, bacteria belonging to the genus *Escherichia, Enterobacter,* or *Pantoea* are preferable.

[0026] The parent strain of *Escherichia* bacteria which can be used to obtain the bacterium of the present invention is not particularly limited, and specifically, bacteria listed in Neidhardt et al. (*Escherichia coli* and *Salmonella Typhimurium,* American Society for Microbiology, Washington D.C., 1029 table 1) may be used. Among these, *Escherichia coli* is preferable. Specific examples of *Escherichia coli* include *Escherichia coli* W3110 strain (ATCC 27325) and *Escherichia coli* MG1655 strain (ATCC 47076), which are derived from the prototype wild-type K12 strain.

[0027] Examples of *Enterobacter* bacteria include *Enterobacter agglomerans* and *Enterobacter aerogenes,* and examples *of Pantoea* bacteria include *Pantoea ananatis.* Recently, *Enterobacter agglomerans* was reclassified in some cases as *Pantoea agglomerans, Pantoea ananatis,* or *Pantoea stewartii* based on an analysis of the nucleotide sequence of 16S rRNA. Therefore, the bacteria of the present invention may belong to the genus *Enterobacter* or *Pantoea,* as long as they are classified in the *Enterobacteriaceae* family. When *Pantoea ananatis* is bred using a genetic engineering technique, *Pantoea ananatis* AJ13355 (FERM BP-6614), AJ13356 strain (FERM BP-6615), AJ13601 (FERM BP-7207),

and derivatives thereof may be used. These strains were identified and deposited as *Enterobacter agglomerans* at the time of isolation, but then reclassified as *Pantoea ananatis* based on an analysis of the nucleotide sequence of 16S rRNA.

[0028] Hereinafter, methods for imparting an L-amino acid-producing ability to such a bacterium as described above will be described.

In order to impart an L-amino acid-producing ability, methods may be used which are typically used in conventional breeding of amino acid-producing bacteria such as a coryneform bacterium or an *Escherichia* bacterium. Such methods include acquisition of nutrient-auxotrophic mutant strains, analogue-resistant strains, and metabolic regulation mutant strains, and breeding recombinant strains having enhanced expression of genes encoding L-amino acid biosynthetic enzymes (Amino Acid Fermentation, Japan Scientific Societies Press, first edition publication: May 30, 1986, p.77 to 100). Nutrient-auxotrophic mutations, analogue-resistant mutations, and metabolic regulation mutations may be imparted singly or in combination during the breeding of L-amino acid-producing bacteria. Furthermore, activities of one or more of the L-amino acid biosynthetic enzymes may be enhanced. Even furthermore, nutrient-auxotrophic mutations, analogue resistant mutations, and metabolic regulation mutations may be imparted in combination with enhancing the activities of the L-amino acid biosynthetic enzymes.

[0029] Nutrient-auxotrophic mutant strains, analogue-resistant strains, and metabolic regulation mutant strains that have an L-amino acid-producing ability can be obtained as follows. That is, a parent strain or a wild-type strain is subjected to a general mutation treatment, i.e., irradiation with X-ray or ultraviolet ray, or a subjected to an agent such as N-methyl-N'-nitro-N-nitrosoguanidine. The strain that exhibits nutrient-auxotrophy, analogue-resistance, or a metabolic regulation mutation and has an L-amino acid-producing ability is selected from the mutated strains.

In addition, an L-amino acid-producing bacterium can be obtained by enhancing an activity of an L-amino acid biosynthetic enzyme via genetic recombination. However, the , L-amino acid-producing ability may be a native property to the wild-type bacterium. Furthermore, as described below, the L-amino acid-producing ability may be imparted by enhancing the expression of the phosphotransacetylase gene.

[0030] Hereinafter, methods of imparting an L-glutamic acid-producing ability and bacteria imparted with L-glutamic acid-producing ability will be described as examples.

In order to impart and/or enhance L-glutamic acid-producing ability, the expression of a gene encoding an enzyme involved in L-glutamic acid biosynthesis can be enhanced. Enzymes involved in L-glutamic acid biosynthesis include glutamate dehydrogenase, glutamine synthetase, glutamate synthase, isocitrate dehydrogenase, aconitate hydratase, citrate synthase, phosphoenolpyruvate carboxylase, pyruvate carboxylase, pyruvate dehydrogenase, pyruvate kinase, phosphoenolpyruvate synthase, enolase, phosphoglyceromutase, phosphoglycerate kinase, glyceraldehyde-3-phosphate dehydrogenase, triosephosphate isomerase, fructosebisphosphate aldolase, phosphofructokinase, and glucose-phosphate isomerase.

[0031] Gene expression can be enhanced by transforming a bacterium with a plasmid containing the desired gene and sequences required for replication of the plasmid, integrating the gene by homologous recombination, conjugation, gene transfer, or the like into the chromosome of the bacterium, or introducing a mutation into the promoter region of the gene (WO 95/34672).

[0032] When transforming the desired gene into a bacterium using a plasmid or integrating the gene into the chromosome, a promoter for expressing the gene may be any promoter as long as it functions in the chosen host bacterium, and it may be the native promoter of the gene or a heterologous promoter.

The expression level of a gene can be increased by selecting a promoter which is known to be strong in the chosen host bacterium, or by modifying the -35 region or the -10 region of the promoter to be close to a consensus sequence. Bacteria modified to have enhanced expression of a citrate synthase gene, an isocitrate dehydrogenase, a pyruvate dehydrogenase gene and/or a glutamate dehydrogenase gene include those described in WO 00/18935 and EP 1010755 A.

[0033] Imparting an L-glutamic acid-producing ability can be performed by decreasing or eliminating an activity of an enzyme that catalyzes a reaction which branches off from the L-glutamic acid biosynthetic pathway and lead to a compound other than L-glutamic acid. Such enzymes include isocitrate lyase, α-ketoglutarate dehydrogenase, acetohydroxyacid synthase, acetolactate synthase, formate acetyltransferase, lactate dehydrogenase, glutamate decarboxylase, and 1-pyrroline dehydrogenase.

[0034] For example, α-ketoglutarate dehydrogenase activity can be decreased by using the sucA (odhA) gene that encodes the Elo subunit of α-ketoglutarate dehydrogenase. Strains having decreased α-ketoglutarate dehydrogenase activity are shown below.

Brevibacterium lactofermentum ΔS strain (WO 95/34672)
Brevibacterium lactofermentum AJ12821 (FERM BP-4172; FR 9401748 A)
Brevibacterium flavum AJ12822 (FERM BP-4173; FR 9401748 A)
Corynebacterium glutamicum AJ12823 (FERM BP-4174; FR 9401748 A)
Pantoea ananatis AJ13601 (FERM BP-7207)
Klebsiella planticola AJ13410 strain (FERM BP-6617)

Pantoea ananatis AJ13355 (FERM BP-6614)

[0035] To decrease or eliminate the activity of such an enzyme, a mutation may be introduced into the gene encoding the enzyme on the chromosome using a general mutation treatment. For example, the gene may be disrupted by gene recombination, or by modifying an expression regulatory sequence such as a promoter and a Shine-Dalgarno (SD) sequence so that the gene expression or translation is decreased. In addition, amino acid substitution (missense mutation), a stop codon (nonsense mutation), a frameshift mutation that adds/deletes one or two nucleotides into a region encoding the enzymes on the chromosome may be introduced, or the gene or a part thereof may be deleted (Journal of Biological Chemistry 272: 8611-8617 (1997). Also, the activity of the enzyme may also be decreased or eliminated by replacing the wild-type gene on the chromosome with a mutant gene in which the coding region is deleted by homologous recombination, or by introducing a transposon or an IS factor into the gene.

[0036] For example, the following method can be used to introduce a mutation that decreases or eliminates the activity of the above-described enzyme by homologous recombination. A mutant gene is prepared by modifying a partial sequence of a target gene so that the encoded enzyme is not produced, and a host bacterium is transformed with the DNA containing the mutant gene. This causes recombination between the mutant gene and the gene on the chromosome, thereby replacing the gene on the chromosome with the mutant gene. Such site-specific mutagenesis by gene substitution using homologous recombination is known, and can be achieved by using a linear DNA or a plasmid containing a temperature-sensitive replication origin (US Patent 6303383 or JP 05-007491 A). Also, site-specific mutagenesis by gene substitution using homologous recombination may also be achieved by using a plasmid which is unable to replicate in the chosen host bacterium.

[0037] Examples of temperature-sensitive plasmids in coryneform bacteria include p48K, pSFKT2 (US Patent 6303383), and pHSC4 (FR 1992-2667875 A and JP 05-7491 A). These plasmids can autonomously replicate at 25°C but cannot autonomously replicate at 37°C. *Escherichia coli* AJ12571 transformed with pHSC4 was deposited at the National Institute of Bioscience and Human-Technology, Agency of Industrial Science and Technology (currently, International Patent Organism Depositary, National Institute of Advanced Industrial Science and Technology, Central 6, 1-1-1 Higashi, Tsukuba, Ibaraki 305-5466, Japan) on October 11, 1990 and given an accession number of FERM P-11763, and then converted to an international deposit under the provisions of Budapest Treaty on August 26, 1991 and given an accession number of FERM BP-3524.

[0038] Moreover, L-glutamic acid-producing ability may be imparted to a host bacterium by amplifying the yggB gene (NCgl 1221;NP_600492. [gi:19552490]; SEQ ID NO: 32) or by introducing a mutant yggB gene containing a mutation in its coding region (WO2006/070944). The following gene may be used as the mutant yggB gene.

(1) Mutation in the C-terminal region

[0039] This mutation is introduced into the region encoding the amino acid sequence of amino acids 419-533 of SEQ ID NO: 33. This mutation may be any mutation as long as it is introduced into the nucleotide sequence encoding the above-described region, and an insertion mutation including insertion of an insertion sequence (hereinafter, referred to as IS), or a transposon, is preferable. The insertion mutation may result in an amino acid substitution (missense mutation), a frameshift, or a stop codon (nonsense mutation). SEQ ID NOS: 7 and 8 show the nucleotide sequence of the mutant yggB gene containing a transposable element (IS) in the C-terminal region and the amino acid sequence encoded by the mutant gene, respectively.

Meanwhile, examples of the mutation in the C-terminal region include replacing a proline in the sequence of amino acids 419-533 of SEQ ID NO: 33 with another amino acid.

(2) Mutation in a transmembrane region

[0040] The Ygg protein encoded by the yggB gene is predicted to have five transmembrane regions, and these transmembrane regions correspond to amino acids 1-23 (first transmembrane region), 25-47 (second transmembrane region), 62-84 (third transmembrane region), 86-108 (fourth transmembrane region), and 110-132 (fifth transmembrane region) in the wild-type YggB protein (SEQ ID NO: 33). A mutation in the transmembrane regions preferably causes substitution, deletion, addition, insertion, or inversion of one or several amino acids without causing a frameshift or a translation termination.

Examples of a mutation in the transmembrane region include inserting one or several amino acids between the leucine at position 14 and the tryptophan at position 15 in the amino acid sequence of SEQ ID NO: 33, replacing the alanine at position 100 with another amino acid, and replacing the alanine at position 111 with another amino acid.

[0041] As another method for imparting and/or enhancing an L-glutamic acid-producing ability, imparting resistance to an organic acid analogue, a respiratory chain inhibitor, etc. and imparting sensitivity to a cell-wall synthesis inhibitor may be used. Examples of such methods include imparting resistance to monofluoroacetic acid (JP 50-113209 A), imparting resistance to adenine or thymine (JP 57-065198 A), attenuating urease (JP 52-038088 A), imparting resistance

to malonic acid (JP S2-038088 A), imparting resistance to benzopyrones or naphthoquinones (JP 56-1889 A), imparting resistance to HOQNO (JP 56-140895 A), imparting resistance to α-ketomalonic acid (JP 57-2689 A), imparting resistance to guanidine (JP 56-35981 A), and imparting sensitivity to penicillin (JP 4-88994 A).
Specific examples of such resistant bacteria include:

Brevibacterium flavum AJ3949 (FERM BP-2632; JP 50-113209 A)
Corynebacterium glutamicum AJ11628 (FERM P-5736; JP 57-065 98 A)
Brevibacterium flavum AJ11355 (FERM P-5007; JP 56-1889 A)
Corynebacterium glutamicum AJ11368 (FERM P-5020; JP 56-1889 A)
Brevibacterium flavum AJ11217 (FERM P-4318; JP 57-2689 A)
Corynebacterium glutamicum AJ11218 (FERM P-4319; JP 57-2689 A)
Brevibacterium flavum AJ11564 (FERM P-5472; JP 56-140895 A)
Brevibacterium flavum AJ11439 (FERM P-5136; JP 56-35981 A)
Corynebacterium glutamicum H7684 (FE-RM BP-3004; JP 04-88994 A)
Brevibacterium lactofermentum AJ11426 (FERM P-5123; JP 56-048890 A)
Corynebacterium glutamicum AJ11440 (FERM P-5137; JP 56-048890 A)
Brevibacterium lactofermentum AJ11796 (FERM P-6402; JP 58-158192 A)

[0042] Bacteria imparted with an L-glutamine-producing ability include a bacterium having enhanced glutamate dehydrogenase activity, a bacterium having enhanced glutamine synthase (glnA) activity, and a bacterium belonging to the *Enterobacteriaceae* family having a disrupted glutaminase gene (EP 1229121 A and EP 1424398 A). The glutamine synthase activity may be enhanced by disrupting the glutamine adenylyltransferase gene (glnE) or by disrupting the gene encoding the PII regulatory protein (glnB), In addition, a preferable L-glutamine-producing bacterium includes an *Escherichia* bacterium having a mutant glutamine synthase in which the tyrosine at position 397 has been replaced with another amino acid (US Patent Publication 2003-0148474).

[0043] Another method of imparting and/or enhancing an L-glutamine-producing ability includes imparting resistance to 6-diazo-5-oxo-norleucine (JP 03-232497 A), imparting resistance to purine analogue and methionine sulfoxide (JP 61-202694 A), and imparting resistance to α-ketomaleic acid (JP 56-151495 A). Specific examples of a coryneform bacterium having an L-glutamine-producing ability include:

Brevibacterium flavum AJ11573 (FERM P-5492; JP 56-161495 A)
Brevibacterium flavum AJ11576 (FERM BP-10381; JP 56-161495 A)
Brevibacterium flavum AJ12212 (FERM P-8123; JP 61-202694 A)

[0044] Bacteria having an L-proline-producing ability include a bacterium harboring γ-glutamyl kinase which is not subject to feedback inhibition by L-proline and a bacterium in which the L-proline degradation system is attenuated. A method of modifying a bacterium using a DNA encoding γ-glutamyl kinase not subject to feedback inhibition by L-proline is disclosed in Dandekar, A.M., Uratsu, S.L., J. Bacteriol., 170, 12, 5943-5 (1988). Also, a bacterium with an attenuated L-proline degradation system is obtained by introducing a mutation into the proline dehydrogenase gene that results in decreased enzymatic activity. Specific examples of a bacterium having an L-proline-producing ability include *Escherichia coli* NRRL B-12403, NRRL B-12404 (GB 2075056), and VKPM B-8012 (US Patent 2002-0058315). Examples of a mutant plasmid for introducing such a mutation include the mutant plasmid disclosed in DE 3127361 B, and the mutant plasmid disclosed by Bloom et al. (The 15th Miami Winter Symposium, 1983, p.34).

[0045] Preferable L-proline-producing bacteria include *Escherichia coli* 702 (VKPM B-8011), which is resistant to 3,4-dehydroxyproline and azatidine-2-carboxylate, 702ilvA (VKPM B-8012), which is an ilvA-dencient derivative of the 702 strain, and an *E. coli* strain having an enhanced activity of a protein encoded by the b2682 gene, b2683 gene, b1242 gene, or b3434 gene (JP 2002-300874 A).

[0046] Examples of L-arginine-producing bacteria include *Escherichia coli* mutant strains which are resistant to α-methylmethionine, p-fluorophenylalanine, D-arginine, arginine hydroxamate, S-(2-aminoethyl)-cysteine, α-methylserine, β-2-thienylalanine, or sulfaguanidine (JP 56-106598 A). A preferable L-arginine-producing bacterium is *Escherichia coli* 237, which is a mutant strain which is resistant to feedback inhibition by L-arginine and has enhanced N-acetylglutamate synthase activity (RU 2000117677). The strain was deposited at the Russian National Collection of Industrial Microorganisms (VKPM), GNII Genetika on April 10, 2000 and given an accession number of VKPM B-7925, and the deposit was then converted to an international deposit under the provisions of Budapest Treaty on May 18, 2001. *Escherichia coli* 382, which is a derivative of the 237 strain and has improved ability to assimilate acetic acid (JP 2002-017342 A), may be used as an L-arginine-producing bacterium. *Escherichia coli* 382 strain was deposited at the Russian National Collection of Industrial Microorganisms (VKPM) on April 10, 2000 and given an accession number of VKPM B-7926.

[0047] Examples of L-arginine-producing bacteria include bacteria modified to have improved expression of a gene

encoding an enzyme involved in L-arginine biosynthesis. L-arginine biosynthetic enzymes include N-acetylglutamate synthase (arg.A), N-acetylglutamyl-phosphate reductase (argC), ornithine acetyltransferase (argJ), N-acetylglutamate kinase (argB), acetylornithine transaminase (argD), acetylornithine deacetylase (argE), ornithine carbamoyl transferase (argF), argininosuccinate synthase (argG), argininosuccinate lyase (argH), and carbamoylphosphate synthase (carAB). The terms in parentheses following the enzyme names refer to names of the genes encoding these enzymes. For N-acetylglutamate synthase (araA), it is more preferable to use a mutant enzyme wherein the wild-type amino acid sequence at positions 15 to 19 is replaced and as a result, the feedback inhibition by L-arginine has been alleviated (EP 1170361 A).

[0048] The biosynthetic pathways of L-citrulline and L-ornithine are the same as that of L-arginine, and the ability to produce L-citrulline and L-ornithine can be imparted by increasing the enzymatic activities of N-acetylglutamate syntase (argA), N-acetylglutamylphosphate reductase (argC), ornithine acetyltransferase (argJ), N-acetylslutamate kinase (argB), acetylornithine transaminase (argD), and acetylornithine deacetylase (argE).

<1-2> Enhancement of phosphotransacerylase activity

[0049] The bacterium of the present invention can be obtained by modifying the above-described bacterium having an L-amino acid-producing ability so that phosphotransacetylase activity is enhanced. However, L-amino acid-producing ability may be imparted after the bacterium is modified so that the phosphotransacetylase activity is enhanced.
The phrase "modified so that phosphotransacetylase activity is enhanced" includes when the number of phosphotransacetylase molecules per cell increases and when the phosphotransacetylase activity per molecule increases as compared to a wild-type strain or unmodified strain. In the case of a coryneform bacterium, examples of the wild-type strain to be used as a control include *Corynebacterium glutamicum (Brevibacterium lactofermentum)* ATCC13869 and ATCC13032. Examples of a wild-type *Escherichia coli* include *Escherichia coli* W3110 (ATCC 27325) and *Escherichia coli* MG1655 (ATCC 47076), which are derived from the prototype wild-type K-12 strain. Examples of a wild-type *Pantoea ananatis* include *Panroea ananatis* AJ13355 (FERM BP-6614) and AJ13356 (FERM BP-6615).

[0050] In the present invention, phosphotransacetylase (phosphoacetyltransferase) catalyzes the reaction to generate acetyl-CoA and phosphate from acetyl phosphate and CoA or the reverse reaction thereof, and is also referred to as phosphate acetyltransferase or phoshoacetylase (EC 2.3.1.8).
In the present invention, enhanced phosphotransacetylase activity can be determined by the method described in Reinscheid D.J. et al. (Microbiology, 145: 503-513 (1999)).
Enhanced phosphotransacetylase activity may also be confirmed by comparing the mRNA level of the gene encoding phosphotransacetylase to that of a wild-type or unmodified strain. Methods of detecting the expression level of a gene include Northern hybridization and RT-PCR (Molecular cloning (Cold spring Harbor Laboratory Press, Cold spring Harbor (USA), 2001). The phosphotransacetylase activity or gene expression level in the bacterium of the present invention may be at any level as long as it is increased as compared to a wild-type or unmodified strain, and for example, the activity or gene expression preferably increases not less than 1.5-fold, more preferably not less than 2-fold, furthermore preferably not less than 3-fold as compared to a wild-type or unmodified strain.

[0051] The gene encoding phosphotransacetylase (pta gene) of a coryneform bacterium includes the nucleotide sequence of NCg12657 (ATCC13032) registered in Genbank (a complementary strand of 2936506..2937891 of accession NC_003450.3). SEQ ID NOS: 40 and 41 show the nucleotide sequence of the gene and the amino acid sequence encoded by the gene, respectively. Furthermore, nucleotide numbers 1214-2641 of SEQ ID NO: 34 and SEQ ID NO: 35 show the nucleotide sequence of the pta gene of *C. glutamicum* ATCC13869 and the amino acid sequence encoded by the gene, respectively.

[0052] Furthermore, the gene of the present invention may be a homologue of the pta gene which is derived from another bacterium as long as it encodes a protein having the phosphotransacetylase activity. Such a homologue can be searched by BLAST (http://blast.genome.jp/) or the like with reference to the nucleotide sequence of nucleotides 1214 to 2641 of SEQ ID NO: 34 or the nucleotide sequence of SEQ ID NO: 40.

[0053] The nucleotide sequence of the pta gene of the present invention has been identified. Therefore, the region containing the pta gene, or the pta gene and its expression regulatory sequences can be obtained by PCR (PCR: polymerase chain reaction; White, T.J. et al., Trends Genet. 5, 185 (1989)) using primers prepared based on the known nucleotide sequence, for example, using the primers of SEQ ID NOS: 9 and 10, and using the chromosomal DNA of a coryneform bacterium as a template. A homologue of the pta gene derived from another bacterium can be obtained in the same way.

[0054] There may be differences between the nucleotide sequences of pta genes depending on species or strains of the bacteria used to isolate the pta gene. Therefore, the pta gene to be used in the present invention is not limited to the nucleotides 1214 to 2641 of SEQ ID NO: 34 or the nucleotide sequence of SEQ ID NO: 40, but may be a mutant or an artificially modified gene which encodes a protein having the amino acid sequence of SEQ ID NO: 35 or 41 and which may include substitutions, deletions, insertions, or additions of one or several amino acids at one or several positions as long as it encodes a protein having the phosphotransacetylase activity. Although depending on the positions in the

ternary structure of the protein or types of amino acids, the term "one or several" used herein specifically means 1 to 20, preferably 1 to 10, more preferably 1 to 5. Meanwhile, such substitutions, deletions, insertions, additions, or inversions include naturally-occurring mutations (mutants or variants) due to differences in individuals or species of the bacteria harboring the pta gene.

[0055] The above-mentioned substitution is preferably a conservative substitution that does not cause a functional change. The conservative substitutions include a substitution between aromatic amino acids such as substitution among Phe, Trp and Tyr; a substitution between hydrophobic amino acids such as substitution among Leu, Ile and Val; a substitution between polar amino acids such as substitution between Gln and Asn; a substitution between basic amino acids such as substitution among Lys, Arg and His; a substitution between acidic amino acids such as substitution between asp and Glu; a substitution between amino acids having a hydroxyl group such as substitution between Ser and Thr. Specific examples of the conservative substitution include: substitution of Ser or Thr for Ala; substitution of Gln, His, or Lys for Arg; substitution of Glu, Gln, Lys, His or Asp for Asn; substitution of Asn, Glu, or Gln for Asp; substitution of Ser or Ala for Cys; substitution of Asn, Glu, Lys, His, Asp, or Arg for Gln; substitution of Gly, Asn, Gln, Lys, or Asp for Glu; substitution of Pro for Gly; substitution of Asn, Lys, Gln, Arg, or Tyr for His; substitution of Leu, Met, Val, or Phe for Ile; substitution of Ile, Met, Val, or Phe for Leu; substitution of Asn, Glu, Gln, His, or Arg for Lys; substitution of Ile, Leu, Val, or Phe for Met; substitution of Trp, Tyr, Met, Ile, or Leu for Phe; substitution of Thr or Ala for Ser; substitution of Ser or Ala for Thr; substitution of Phe or Tyr for Trp; substitution of His, Phe, or Trp for Tyr; and substitution of Met, Ile, or Leu for Val.

[0056] Furthermore, the pta gene may be a gene that has a nucleotide sequence having at least 80%, preferably at least 90%, more preferably 95%, particularly preferably at least 97% homology to the entire amino acid sequence of SEQ ID NO: 35 or 41, and encodes a protein having the phosphotransacetylase activity. Also, the pta gene may be modified to have a codon that is easily used by the chosen host bacterium because the degeneracy levels of genes are different depending on the host bacterium.

The pta gene may have an elongated or shortened sequence at the N-terminal side and/or C-terminal side as long as it encodes a protein having phosphotransacetylase activity. The length of the amino acid sequence which is elongated or shortened is 50 or less, preferably 20 or less, more preferably 10 or less, particularly preferably 5 or less. More specifically, the pta gene may have an amino acid sequence of SEQ ID NO: 35 or 41 in which an additional 50 to 5 amino acids are present at the N-terminal side and/or at the C-terminal side.

[0057] A homologue of the pta gene can be obtained by modifying the nucleotide sequence of nucleotides 1214 to 2641 of SEQ ID NO: 34 or the nucleotide sequence of SEQ ID NO: 40 so that specific amino acid residues in the protein encoded by the gene are substituted, deleted, inserted, or added, for example, by site-specific mutagenesis. Furthermore, a homologue gene may also be obtained by a known mutation treatment as described below. A homologue gene may be obtained by treating the above-described nucleotide sequence in vitro with hydroxylamine or the like, treating a bacterium harboring the gene, for example, a coryneform bacterium with ultraviolet rays or with a mutagen such as N-methyl-N'-nitro-N-nitrosoguanidine (NTG) or ethylmethanesulfonate (EMS), or by artificially introducing a mutation into the pta gene by gene recombination such as error-prone PCR, DNA shuffling, and StEP-PCR (Firth AE, Patrick WM; Bioinformatics. 2005 Jun 2; Statistics of protein library construction). Determination of the enzymatic activity by the above-described method can confirm whether such a pta gene homolog encodes a protein having the phosphotransacetylase activity.

[0058] Examples of the pta gene include a DNA that hybridizes with a sequence complementary to nucleotides 1214 to 2641 of SEQ ID NO: 34 or the nucleotide sequence of SEQ ID NO: 40, or with a probe that can be prepared from the sequences under stringent conditions and encodes a protein having the phosphotransacetylase activity. The term "stringent conditions" used herein includes conditions under which so-called specific hybrid is formed and non-specific hybrid is not formed. Examples thereof include conditions under which DNAs having high homology of at least 80%, preferably at least 90%, more preferably at least 95%, further more preferably at least 97% hybridize with each other and DNAs having homology of less than 80% do not hybridize with each other; and specifically includes conditions of washing in general Southern hybridization, i.e., conditions including washing at a salt concentration of 1 x SSC, 0.1% SDS, preferably 0.1 x SSC, 0.1% SDS at 60°C, preferably 68°C, once, preferably twice or three times.

[0059] As a probe, a partial sequence of nucleotides 1214 to 2641 of SEQ ID NO: 34 or a partial sequence of the nucleotide sequence of SEQ ID NO: 40 may be used. Such a probe can be prepared by PCR using oligonucleotides prepared based on the nucleotide sequences as primers and a DNA fragment having the nucleotide sequence of nucleotides 1214 to 2641 of SEQ ID NO: 34 or the nucleotide sequence of SEQ ID NO: 40 as a template. For example, when using a DNA fragment having a length of about 300 bp, exemplary washing conditions include hybridization in 2 x SSC, 0.1% SDS at 50°C.

[0060] The expression of the pta gene can be enhanced by increasing the copy number of the pta gene. For example, the expression can be enhanced by preparing a recombinant DNA by ligating a fragment containing the pta gene into a vector that functions in the host bacterium, preferably a multi-copy vector; and then transforming the above-described bacterium having an L-amino acid-producing ability with the recombinant DNA. Alternatively, the expression may be

enhanced by introducing the above-described recombinant DNA into a wild-type bacterium to prepare a transformant, and then imparting an L-amino acid-producing ability to the transformant. Meanwhile, the copy number may be increased by transferring one or more copies of the pta gene on the chromosome. Integration of the pta gene into the chromosome can be confirmed by Southern hybridization using a part of the pta gene as a probe.

[0061] Expression of the pta gene can also be enhanced by modifying an expression regulatory region of the pta gene. For example, it can be achieved by replacing a promoter sequence of the pta gene with a more potent promoter, or modifying a promoter sequence to be close to a consensus sequence (WO 00/18935).

[0062] Hereinafter, methods of constructing a coryneform bacterium modified so that the phosphotransacetylase activity is increased will be shown. These methods can be performed in accordance with the manuals of Molecular cloning (Cold spring Harbor Laboratory Press, Cold spring Harbor (USA), 2001) and the like. Bacteria other than coryneform bacteria may also be modified so that the phosphotransacetylase activity is increased by the same method.

[0063] The expression level of the pta gene can be enhanced by increasing the copy number of the pta gene, and the copy number may be increased by amplifying the pta gene using a plasmid as described below. First, the pta gene is cloned from the chromosome of a coryneform bacterium. The chromosomal DNA may be prepared from a bacterium that serves as a DNA donor by, for example, the method of Saito and Miura (Biochem. Biophys. Acta, 72, 619 (1963), Experiment Manual for Biotechnology, edited by The Society for Biotechnology, Japan, p97-98, Baifukan Co., Ltd., 1992). Oligonucleotides for PCR are synthesized based on the above-described known sequence information, and the pta gene can be amplified by using the synthetic oligonucleotides of SEQ ID NOS: 9 and 10, for example.

[0064] A gene fragment containing the amplified pta gene is ligated to a vector which is autonomously replicable in *Escherichia coli* and/or coryneform bacterium, and the recombinant DNA is introduced into *Escherichia coli.* Examples of a vector which is autonomously replicable in *Escherichia coli* include pUC19, pUC18, pHSG299, pHSG399, pHSG398, RSF1410, pBR322, pACYC184, and pMW219.

[0065] The above-mentioned DNA is introduced into a vector capable of functioning in coryneform bacteria. An example of such a vector includes a plasmid capable of autonomously replicating in coryneform bacteria. Specific examples thereof include pCRY30 described in JP 3-210184 A; pCRY21, pCRY2KE, pCRY2KX, pCRY31, pCRY3KE, and pCRY3KX described in JP 2-72876 A and US Patent 5,185,262; pCRY2 and pCRY3 described in JP 1-191686 A; pAM330 described in JP 58-67679 A; pHM1519 described in JP 58-77895 A; pAJ655, pAJ611, and pAJ1844 described in JP 58-192900 A; pCG1 described in JP 57-134500 A; pCG2 described in JP 58-35197 A; pCG4 and pCG11 described in JP 57-183799 A; and pVK7 described in JP 10-215883 A.

[0066] Furthermore, if a DNA fragment able to autonomously replicate in a coryneform bacterium is excised from one of these plasmids and inserted into the above-described vector for *Escherichia coli,* this vector can be used as a so-called shuttle vector, and it will be autonomously replicable in both *Escherichia coli* and coryneform bacteria.

[0067] These vectors can be obtained from the deposited bacteria shown in the above-described patent documents as follows. Cells collected at the logarithmic growth phase are lysed with lysozyme and SDS, and centrifuged at 30,000 x g, and then polyethylene glycol is added to the supernatant, followed by separation and purification by cesium chloride-ethidium bromide equilibrium gradient centrifugation.

[0068] To prepare a recombinant DNA by ligating the pta gene with a vector that functions in coryneform bacteria, the vector is digested with a restriction enzyme suitable for excising a fragment containing the pta gene. The restriction site can be introduced in advance into a synthetic oligonucleotide for amplifying the pta gene. The ligation is generally performed using a ligase such as T4DNA ligase.

[0069] The recombinant plasmid prepared as described above may be introduced into a coryneform bacterium by any transformation method reported so far. Examples thereof include increasing the permeability of a DNA by treating recipient cells with calcium chloride, which has been reported for *Escherichia coli* K-12 (Mandel, M. and Higa, A., J. Mol. Biol., 53, 159 (1970), and introducing a DNA into competent cells prepared from cells at the proliferation stage, which has been reported for *Bacillus subtilis* (Duncan, C.H., Wilson, G.A and Young, F.E, Gene, 1, 153 (1977)). Alternatively, preparing protoplast-like cells or spheroplast-like cells of a DNA-recipient bacterium, which can easily incorporate a recombinant DNA, and introducing a recombinant DNA into these cells may also be used (Chang, S. and Choen, S.N., Mol. Gen. Genet., 168, 111 (1979); Bibb, M.J., Ward, J.M. and Hopwood, O.A., Nature, 274, 398 (1978); Hinnen, A., Hicks, J.B. and Fink, GR., Proc. Natl. Acad. Sci. USA, 75 1929 (1978)). A coryneform bacterium can also be transformed by the electric pulse method (JP 02-207791 A) or the conjugation transfer method (Biotechnology (NY). 1991 Jan; 9(1): 84-7).

[0070] The copy number of the pta gene can be increased by introducing multiple copies of the pta gene into the chromosomal DNA of a coryneform bacterium. In order to introduce multiple copies of the pta gene into the chromosomal DNA of a coryneform bacterium, homologous recombination is performed using a sequence which is present on a chromosomal DNA in multiple copies. Repetitive DNA or an inverted repeat existing at the end of a transposable element can be used as a sequence which is present on a chromosomal DNA in multiple copies. Alternatively, as disclosed in JP 02-109985 A, multiple copies of the pta gene may be introduced by inserting the gene into a transposon, to thereby transfer multiple copies of the gene into a chromosomal DNA (JP 02-109985 A, JP 07-107976 A, and Mol. Gen. Genet.,

245, 397-405 (1994), Plasmid. 2000 Nov; 44(3):285-91).

[0071] Furthermore, the pta gene may be amplified on the chromosome by inserting the pta gene into a plasmid having a replication origin which cannot replicate in the host or a plasmid having a replication origin which cannot replicate in the host and is capable of conjugation transfer. Examples of such a plasmids include pSUP301 (Simo et al., Bio/Technology 1, 784-791 (1983)), pK18mob, or pK19mob (Schaefer et al., Gene 145, 69-73 (1994)), pGEM-T (Promega corporation, Madison, WI, USA), pCR2.1-TOPO (Shuman (1994). Journal of Biological Chemistry 269: 32678-84; US Patent 5487993), pCR(R)Blunt (Invitrogen, Groningen, Netherlands; Bernard et al., Journal of Molecular Biology, 234: 534-541 (1993)), pEM1 (Schrumpf et al., 1991, Journal of Bacteriology 173: 4510-4516), and pBGS8 (spratt et al., 1986, Gene, 41: 337-342). A plasmid vector containing the pta gene is transferred to a coryneform bacterium by conjugation or transformation. A method of the conjugation is described, for example, by Schaefer et al. (Applied and Environmental Microbiology 60, 756-759 (1994)). Methods of the transformation are described, for example, by Theirbach et al. (Applied Microbiology and Biotechnology 29, 356-362 (1988)), Dunican and Shivinan (Bio/Technology 7, 1067-1070 (1989)), and Tauch et al. (FEMS Microbiological Letters 123, 343-347 (1994)).

[0072] Expression of the pta gene may also be increased by replacing an expression regulatory sequence such as the promoter of the pta gene on the chromosomal DNA or on a plasmid with a more potent promoter, modifying a factor involved in the regulation of expression of the pta gene, for example, an operator or a repressor, or ligating a more potent terminator (Hamilton et al.; Journal of Bacterology 171: 4617-4622). Examples of known potent promoters include the lac promoter, trp promoter, trc promoter, and PS2 promoter. Methods of evaluating promoter strength and examples of potent promoters are described by Goldstein et al. (Prokaryotic promoters in biotechnology. Biotechnol. Annu. Rev., 1995, 1. 105-128). As disclosed in WO 00/18935, a promoter may be made stronger by introducing a substitution of several nucleotides into the promoter region of the target gene so that the promoter becomes close to a consensus sequence. For example, the -35 region may be changed into TTGACA or TTGCCA, while the -10 region may be changed into TATAAT or TATAAC. In addition, it is known that translation efficiency of mRNA is significantly affected by substituting several nucleotides in a spacer sequence between the ribosome binding site (RBS) and the translation initiation codon, in particular, the sequence immediately upstream of the translation initiation codon, and in this way, the spacer sequence may be modified.

[0073] An example of an upstream region of the pta gene includes nucleotides 1 to 1213 of SEQ ID NO: 34. An expression regulatory sequence such as a promoter of the pta gene can be determined using a promoter search vector or gene-finding software such as GENETYX. The above-described promoter substitution or modification enhances the expression of the pta gene. The expression regulatory sequence may also be substituted by using a temperature-sensitive plasmid. The modification of an expression regulatory sequence may be combined with increasing the copy number of the pta gene.

Furthermore, the expression may also be increased by extending the retention time of the mRNA or by preventing degradation of the PTA protein in the cell.

[0074] Also, there is a binding site for the RamB protein which negatively regulates the expression of the pta gene. This binding site is located upstream of the pta gene in coryneform bacterium, and thus, the RamB protein-binding site may be modified to increase the pta activity. The RamB protein-binding sites at the upstream of a pta gene have been reported in J Bacteriol. 2004 May; 186 (9): 2798-2809, and are predicted to include AAAACTTTGCAAA starting at position -87 and AAAACTTTGCAAA starting at position -203 as counted from the translation initiation codon of the pta gene (the sequences of these RamB protein-binding sites correspond to nucleotides 1000 to 1012 and 1115 to 1127 of SEQ ID NO: 34, respectively). A RamB protein-binding sequence is reported to have a consensus sequence of A(A/G)AACTTTGCAAA and is located upstream of the aceA gene which encodes isocitrate lyase, the aceB gene which encodes malate synthase, and the pta-ack operon, and the expression of these genes is induced in the presence of a high concentration of acetic acid. Therefore, modification of the conserved sequence can prevent the decrease in expression of the pta gene due to binding of RamB, and examples of a modification of the sequence include replacing an AT base pair with a GC base pair and replacing a GC base pair with an AT base pair, specifically include replacing AAAACTTT-GCAAA with aaaacGAGgcGaG or aaaacGAGgcaaa.

Whether the pta gene is modified so that it is negatively regulated by RamB can be confirmed by examining if the pta gene is constitutively expressed in the absence of acetic acid (for example, only glucose is present as a carbon source) (J Bacteriol. 2004 May; 186 (9): 2798-809).

[0075] The phosphotransacetylase activity may also be increased by modifying a regulator protein that regulates the expression of the pta gene. An example of a regulator for the pta gene includes the above-described RamB protein since it negatively controls the expression of the pta gene. The ramB gene of coryneform bacterium has been registered in Genbank. For example, the ramB gene of *glutamicum* ATCC13032 strain is registered under an accession No. NC_006958.1:390784..392208. SEQ ID NOS: 42 and 43 show the sequence of the ramB gene and the amino acid sequence encoded by the ramB gene, respectively. Meanwhile, SEQ ID NO: 36 and 37 show the ramB gene of *C. glutamicum* ATCC13869 strain and the amino acid sequence encoded by the ramB gene, respectively. The RamB gene may be a DNA which hybridizes with a complementary strand of the nucleotide sequence of SEQ ID NO: 36 or 42 or with a probe

prepared therefrom under stringent conditions, and encodes a protein that binds to the RamB-binding sites to represses the expression of the pta gene. Here, the stringent conditions are as mentioned above.

[0076] Expression of the ramB gene may be decreased to enhance the expression of the pta gene. For example, a mutation that decreases or eliminates the expression of the ramB gene may be introduced by a gene recombination technique as follows. A partial sequence of the ramB gene is mutated so that the RamB protein is not produced, and a bacterium is transformed with a DNA comprising the mutant ramB gene to cause recombination between the mutant ramB gene and a wild-type ramB gene on the chromosome, resulting in replacement of the wild-type ramB gene on the chromosome with the mutant ramB gene.

[0077] The wild-type ramB gene on the chromosome of a host bacterium can be replaced by the mutant ramB gene by the following procedures, for example. First, a plasmid for recombination is prepared by inserting into a plasmid vector a temperature-sensitive replication origin, the mutant ramB gene, a sacB gene encoding a levansucrase, and a marker gene that exhibits resistance to an antibiotics, such as chloramphenicol.

[0078] Herein, the sac B gene encodes a levansucrase, and is used for effectively selecting a strain in which a vector portion has been cured from the chromosome (Schafer, A. et al. Gene 145 (1994) 69-73). When a levansucrase is expressed in a bacterium, the bacterium cannot grow because levan generated by assimilation of sucrose lethally effects the bacterium. If a strain in which the sacB gene-carrying vector remains on the chromosome is cultured on a sucrose-containing plate, it cannot grow. Therefore, only a strain in which the vector has been cured from the chromosome can be selected on a sucrose-containing plate.

[0079] Examples of a sacB gene and homologues thereof include the following:

Bacillus subtilis: sacB GenBank Accession No. X02730 (WO 2005/113745)
Bacillus amyloliquefaciens: sacB GenBank Accession No. X52988
Zymomonas mobilis: sacB GenBank Accession No. L33402
Bacillus stearothermophilus: surB GenBank Accession No. U34874
Lactobacillus sanfranciscensis: frfA GenBankAccession No. AJ508391
Acetobacter xylinus: lsxA GenBank Accession No. AB034152
Gluconacetobacter diazotrophicus: lsdA GenBank Accession No. L41732

[0080] The transformant obtained by introducing a plasmid containing the sacB gene and the mutant ramB gene is cultured at an appropriate temperature for the functioning of the temperature-sensitive replication origin (25°C). This strain is cultured at a high temperature (for example, 34°C) so that the temperature-sensitive replication origin cannot function, resulting in the curing of the temperature-sensitive plasmid, and then this strain is cultured on a plate containing an antibiotic. Since the temperature-sensitive plasmid cannot replicate at the high temperature, a strain which has been cured of the plasmid cannot grow on a plate containing an antibiotic, but a strain in which recombination occurs between the mutant ramB gene on the plasmid and the wild-type ramB gene on the chromosome can grow, and colonies appear.

[0081] In a strain containing the mutant ramB gene integrated into the chromosomal DNA, recombination is caused between the mutant ramB gene and the wild-type ramB gene native to the chromosome, and the fusion genes of the wild-type ramB gene and the mutant ramB gene are inserted into the chromosome so that the other portions of the recombinant DNA (vector segment, temperature sensitive replication origin and antibiotic resistance marker) are present between the fusion genes.

[0082] Then, in order to leave only the mutant ramB gene on the chromosome, one copy of the ramB gene is eliminated together with the vector segment (including the temperature-sensitive replication origin and the antibiotics resistance marker) from the chromosomal DNA. In this case, the wild-type ramB gene is left on the chromosomal DNA and the mutant ramB gene is excised from the chromosomal DNA, or to the contrary, the mutant ramB gene is left on the chromosomal DNA and the wild-type ramB gene is excised from the chromosome DNA. In both cases, the excised DNA is maintained in the host bacterium when the host bacterium is cultured at a temperature which allows the temperature-sensitive replication origin to function. Then, the gene on the plasmid is cured from the cells along with the plasmid by culturing the bacterium at a temperature which does not allow the temperature-sensitive replication origin to function. In the case of the sacB gene, strains from which the plasmid is cured can be efficiently obtained by culturing the bacterium in a sucrose-containing medium. Strains in which the wild-type ramB gene is replaced with the mutant ramB gene can be obtained by selecting strains which harbor the mutant ramB gene from the plasmid-cured strains.

[0083] The bacterium of the present invention may be modified so that the activity/activities of D-xylose 5-phosphate-phosphoketolase and/or fructose 6-phosphate phosphoketolase is/are increased in addition to enhancing the phosphotransacetylase activity.

At least one of the D-xylose 5-phosphate-phosphoketolase activity and fructose 6-phosphate phosphoketolase activity may be increased. In the present description, D-xylose 5-phosphate-phosphoketolase and fructose 6-phosphate phosphoketolase are sometimes collectively referred to as phosphoketolase.

The D-xylose 5-phosphate-phosphoketolase activity means an activity which results in conversion of xylose-5-phosphate

into glycelaldehyde-3-phosphate and acetyl phosphate by consumption of phosphoric acid, with concomitant release of one molecule of H_2O. This activity can be determined by the method described by Goldberg, M. et al. (Methods Enzymol., 9, 515-520 (1996) or by the method described by L. Meile (J. Bacteriol. (2001) 183; 2929-2936).

Meanwhile, the fructose 6-phosphate phosphoketolase activity means an activity which results in conversion of fructose 6-phosphate into erythrose-4-phosphate and acetyl phosphate by consuming phosphoric acid, with concomitant release of one molecule of H_2O. This activity can be determined by the method described by Racker, E (Methods Enzymol., 5, 276-280 (1962)) or by the method described by L. Meile (J. Bacteriol. (2001) 183; 2929-2936).

[0084] The phosphoketolase activity is enhanced preferably not less than 1.5-fold, more preferably not less than 2-fold, particularly preferably not less than 3-fold as compared to an unmodified strain or a wild-type strain.

As is the case when enhancing phosphotransacetylase activity as described above, the phosphoketolase activity can be enhanced by increasing the copy number of the gene encoding phosphoketolase or modifying a promoter of the gene encoding phosphoketolase.

The phosphoketolase gene of the host bacterium may be amplified, and the phosphoketolase activity may be imparted by introducing a heterologous gene if the host bacterium has no phosphoketolase activity.

A gene encoding D-xylose 5-phosphate-phosphoketolase can be obtained by PCR using as a template the chromosomal DNA of a bacterium having an enzyme activity of D-xylose 5-phosphate-phosphoketolase. Examples of such a bacterium include bacteria such as lactic bacteria, methanol-assimilating bacteria, methane-assimilating bacteria, bacteria belonging to the genus *Streptococcus, Acetobacter, Bifidobacterium, Lactobacillus, Thiobacillus, Methylococcus, Butyrivibrio,* or *Fibrobactor;* and yeasts belong to the genus *Candida, Rhodotorula, Rhodosporidium, Pichia, Yarrowia, Hansenula, Kluyveromyces, Saccharomyces, Trichosporon, Wingea,* or the like.

The gene encoding fructose 6-phosphate phosphoketolase can be obtained by PCR using as a template the chromosomal DNA of a bacterium having an enzymatic activity of fructose 6-phosphate phosphoketolase. Examples of such a bacterium include bacteria belonging to the genus *Acetobacter, Bifidobacterium, Chlorobium, Brucella, Methylococcus,* or *Gardnerella;* and yeasts belong to the genus *Candida, Rhodotorula, Saccharomyces,* or the like.

[0085] A specific example of the gene encoding D-xylose 5-phosphate phosphoketolase is the xpkA gene derived from *Lactobacillus pentosus* MD363. The nucleotide sequence thereof is registered with an accession number of AJ309011 (Posthuma, C.C. et al, Appl. Environ. Microbiol., 68(2), 831-7(2002))(SEQ ID NO: 52) in the EMBL/GenBank database. The xpkA gene may hybridize with a complementary strand of the nucleotide sequence of SEQ ID NO: 52 or with a probe prepared therefrom under stringent conditions, and which encodes a protein that has D-xylose 5-phosphate phosphoketolase activity.

The xpk1 gene derived from *Lactobacillus plantarum* can also be used. The nucleotide sequence thereof is registered with an accession number of NC_004567 Region (complement of 2362936..2365302) (Kleerebezem, M., et al, Proc. Natl. Acad. Sci. U.S.A. 100 (4), 1990-1995 (2003))(SEQ ID NO: 54) in the EMBL/GenBank database. The xpk1 gene may hybridize with a complementary strand of the nucleotide sequence of SEQ ID NO: 54 or with a probe prepared therefrom under stringent conditions and encodes a protein that has D-xylose 5-phosphate phosphoketolase activity.

In addition, examples of homologs of these genes include a gene of *Lactobacillus plantarum* which is registered as GenBank Accession No. NC_004567 (complement of 3169067-3171478), a gene of *Streptococcus agalactiae* encoding the amino acid sequence of GenBank Accession No. NP_736274, a gene of *Lactococcus lactis* subsp. *Lactis* encoding the amino acid sequence of GenBank Accession No. NP_267658, a gene of *Lactobacillus johnsonii* which is registered as GenBank Accession No. NC_005362 (696462..698867), and a gene of *Lactobacillus acidophilus* encoding the amino acid sequence of GenBank Accession No. YP_193510.

Other examples of fructose 6-phosphate phosphoketolase gene and/or D-xylose 5-phosphate phosphoketolase gene are disclosed in WO2006/016705.

[0086] A gene encoding a protein having the activities of both D-xylose 5-phosphate phosphoketolase and fructose 6-phosphate phosphoketolase can also be used. Examples of such a gene include the xfp gene of *Bifidobacterium animalis.* The nucleotide sequence thereof is registered as accession number of AJ293946 (Meile, L. et al, J. Bacteriol., 183(9), 2929-36 (2001))(SEQ ID NO: 56) in the EMBL/GenBank database. The xfp gene may hybridize with a complementary strand of the nucleotide sequence of SEQ ID NO: 56 or with a probe prepared therefrom under stringent conditions, and which encodes a protein that has D-xylose 5-phosphate phosphoketolase activity and fructose 6-phosphate phosphoketolase activity.

In addition, examples of a homolog of the xfp gene include a gene isolated from *Bifidobacterium longum* which encodes the amino acid sequence of GenBank Accession No. NP_696135, a gene isolated from *Chlorobium tepidum* which encodes the amino acid sequence of GenBank Accession No. NP_662409, a gene isolated from *Brucella suis* which encodes the amino acid sequence of GenBank Accession No. NP_699578, and a gene isolated from *Brucella abortus* which encodes the amino acid sequence of GenBank Accession No. YP_223570.

[0087] The bacterium of the present invention may be modified so that pyruvate carboxylase activity is increased in addition to enhancing phosphotransacetylase activity, or in addition to enhancing the activities of phosphotransacetylase and D-xylose 5-phosphate-phosphoketolase and/or fructose 6-phosphate phosphoketolase.

[0088] Examples of the pyruvate carboxylase gene include the pyc gene derived from coryneform bacterium and the pyc gene derived from *Bacillus* bacterium and specific examples thereof include the pyc gene derived from *C. glutamicum* ATCC13032 strain (GenBank Accession No. NCg10659: SEQ ID NO: 64) and the pyc gene derived from *B. subtilis* (EP 1092776). The pyruvate carboxylase gene may hybridize with a complementary strand of the nucleotide sequence of SEQ ID NO: 64 or with a probe prepared therefrom under stringent conditions, and which encodes a protein that has pyruvate carboxylase activity.

[0089] The bacterium of the present invention may also be modified so that phosphoenolpyruvate carboxylase activity is increased in addition to enhancing the phosphotransacetylase activity or in addition to the enhancing the activities phosphotransacetylase, D-xylose 5-phosphate-phosphoketolase and/or fructose 6-phosphate phosphoketolase. Examples of a phosphoenolpyruvate carboxylase gene include the ppc gene derived from coryneform bacterium and the ppc gene derived from *Escherichia* bacterium and specific examples thereof include the ppc gene from *C. glutamicum* ATCC13032 strain (GenBank Accession No. NCgl1523: SEQ ID NO: 62) and the ppc gene derived from *E. coli* MG1655 strain (GenBank Accession No. NP_418391). The phosphoenolpyruvate carboxylase gene may hybridize with a complementary strand of the nucleotide sequence of SEQ ID NO: 62 or with a probe prepared therefrom under stringent conditions, and which encodes a protein that has phosphoenolpyruvate carboxylase activity. Furthermore, since some kind of phosphoenolpyruvate carboxylase is sensitive to feedback inhibition by aspartic acid, it is preferably modified so it is resistant to the feedback inhibition by aspartic acid (EP0723011).

<2> Method of producing L-amino acid

[0090] An L-amino acid can be produced by culturing a bacterium obtained as described above in a medium to produce and accumulate an L-amino acid in the medium or the bacterial cells and collecting the L-amino acid from the medium or from the bacterial cells.

[0091] The medium can be a general medium containing a carbon source, nitrogen source, inorganic salts, and if necessary, organic trace nutrients such as an amino acid or a vitamin. Either a synthetic or a natural medium may be used. The carbon source and nitrogen source may be any substance as long as it can be utilized by the chosen strain.

[0092] Sugars such as glucose, glycerol, fructose, sucrose, maltose, mannose, galactose, starch hydrolysate, and molasses may be used as the carbon source, and an organic acid such as acetic acid and citric acid and an alcohol such as ethanol may also be used singly or in combination with another carbon source. As a nitrogen source, an ammonium salt such as ammonia, ammonium sulfate, ammonium carbonate, ammonium chloride, ammonium phosphate, or ammonium acetate, or a nitrate salt may be used. As organic trace nutrients, an amino acid, a vitamin, a fatty acid, and a nucleic acid, as well as peptone, casamino acid, yeast extracts, soybean hydrolysate containing these nutrients may be used, and in the case of using a nutrient-auxotrophic mutant strain that requires an amino acid or the like for growth, the required nutrients are supplemented. A phosphate salt, a magnesium salt, a calcium salt, an iron salt, a manganese salt, or the like may be used as the inorganic salt.

[0093] The culture is preferably performed with aeration while adjusting the fermentation temperature to 20 to 45°C and the pH to 3-9. If the pH is lowered during the culture, the medium is neutralized by adding calcium carbonate or an alkaline such as ammonia gas or the like. The culture is performed under such conditions preferably for about 10 to 120 hours, thereby, a large amount of L-amino acid is able to accumulate in the culture medium.

[0094] When producing L-glutamic acid, the culture may be performed using a liquid medium which is adjusted to the appropriate conditions for producing L-glutamic acid, while at the same time precipitating L-glutamic acid into the medium. Examples of such conditions include a pH range of 5.0 to 4.0, preferably of pH 4.5 to 4.0, more preferably of pH 4.3 to 4.0, particularly preferably of pH 4.0 (EP 1078989 A).

[0095] The L-amino acid can be collected from the culture medium by known collection methods. For example, the L-amino acid can be collected by removing bacterial cells from the culture medium, followed by concentration and crystallization, ion exchange chromatography, or the like. When culturing under conditions for precipitating L-glutamic acid, L-glutamic acid can be collected by centrifugation, filtration, or the like. In this case, L-glutamic acid which is dissolved in the medium may also be crystallized and then collected.

EXAMPLES

[0096] Hereinafter, the present invention will be specifically described by referring to the following non-limiting examples.

Example 1: Construction of the pta gene-amplified strain of ATCC13869

[0097] A strain in which the sucA gene is disrupted, and a mutant yggB is introduced, was used as a parental strain for amplifying a pta gene. This strain can be constructed by the following methods.

(1-1) Construction of a sucA-disrupted strain

[0098] A sucA gene-disrupted strain (ATCC13869ΔsucA) was constructed as follows.
Disruption of the sucA gene encoding the Elo subunit of α-ketoglutarate dehydrogenase was performed using pBS3, which carries the sacB gene which encodes levansucrase, according to the method described in WO 2005/113745 and 2005/113744.
A gene fragment which is derived from the *C. glutamicum* ATCC13869 strain and in which the ORF of the sucA gene was deleted, was obtained by the overlap PCR method using synthetic DNAs designed based on the known nucleotide sequence of the sucA gene (SEQ ID NO: 30) of *C. glutamicum* ATCC13032 (GenBank Database Accession No. NC_003450) as primers. Specifically, PCR was performed by a conventional method using a chromosomal DNA of *C. glutamicum* ATCC13869 strain as a template and using synthetic DNAs of SEQ ID NOS: 1 and 2 as primers, to thereby obtain an amplified product corresponding to the N-terminal region of the sucA gene. To obtain an amplified product corresponding to the C-terminal region of the sucA gene, PCR was performed using the chromosomal DNA of the *C. glutamicum* ATCC13869 strain as a template, and using synthetic DNAs of SEQ ID NOS: 3 and 4 as primers. SEQ ID NOS: 2 and 3 are complementary to each other, and are designed to amplify the sucA gene which has entire ORF deleted. Subsequently, to obtain a sucA gene fragment wherein the internal sequence is deleted, the above-described amplified products each corresponding to the N-terminal region and the C-terminal region of the sucA gene were mixed in approximately equal molar amounts, and PCR was performed using the mixture as templates and using synthetic DNAs of SEQ ID NOS: 5 and 6 as primers, to thereby obtain a sucA gene-amplified product into which the deletion mutation was introduced. The PCR product was purified by a conventional method, and then digested with BamHI, followed by insertion into the BamHI site of the above-described pBS3. The DNA was used to transform competent cells of *Escherichia coli* JM109 (Takara Bio Inc.), and the transformed cells were cultured overnight on an LB plate containing 100 μM of IPTG, 40 μg/ml of X-Gal, and 25 μg/ml of kanamycin (Km). Single colonies of transformants were isolated by selecting the white colonies which appeared. Plasmids were extracted from these transformants, and those containing the desired PCR product were named pBS3ΔsucA.

[0099] The pBS3ΔsucA obtained in above (A) does not contain a region that allows autonomous replication in coryneform bacteria. Therefore, when a coryneform bacterium is transformed with the plasmid, only transformants containing the plasmid integrated into the chromosome by homologous recombination will appear at low frequency. The *C. glutamicum* ATCC13869 strain was transformed with a high concentration of the plasmid pBS3ΔsucA by the electric pulse method, and the cells were applied to a CM-Dex plate (5 g/L of glucose, 10 g/L of polypeptone, 10 g/L of yeast extract, 1 g/L of KH_2PO_4, 0.4 g/L of $MgSO_4 \cdot 7H_2O$, 0.01 g/L of $FeSO_4 \cdot 7H_2O$, 0.01 g/L, of $MnSO_4 \cdot 7H_2O$, 3 g/L of urea, 1.2 g/L of soybean hydrolysate, 10 μg/L of biotin, 15 g/L of agar, pH was adjusted to 7.5 with NaOH) containing 25 μg/ml of kanamycin, and cultured at 31.5°C for about 30 hours. In the cells which grew on this medium, the kanamycin-resistant gene and the sacB gene from the plasmid were integrated into the chromosome by homologous recombination between the sucA gene of the plasmid and the sucA gene on the chromosome of the ATCC13869 strain.
Subsequently, the single-crossover recombinants which were obtained were cultured at 31.5°C overnight in CM-Dex liquid medium containing no kanamycin, and after suitable dilution, the cells were applied to a 10% sucrose-containing Dex-S10 plate (100 g/L of sucrose, 10 g/L of polypeptone, 10 g/L of yeast extract, 1 g/L of KH_2PO_4, 0.4 of g/L $MgSO_4 \cdot 7H_2O$, 0.01 g/L of $FeSO_4 \cdot 7H_2O$, 0.01 g/L of $MnSO_4 \cdot 4H_2O$, 3 g/L of urea, 1.2 g/L of soybean hydrolysate, 10 μg/L of biotin, 15 g/L of agar, pH was adjusted to 7.5 with KOH) containing no kanamycin, and cultured at 31.5°C for about 30 hours. As a result, strains which were sucrose-insensitive due to the curing of the sacB gene by the second homologous recombination were obtained.
The strains thus obtained include one in which the sucA gene was replaced by the mutant gene derived from pBS3ΔsucA, and one in which the sucA gene has reverted to the wild-type gene. Whether the sucA gene is the mutant gene or the wild-type gene can be easily confirmed by directly subjecting the bacterial cells obtained by culture on a Dex-S10 plate to PCR, and detecting the sucA gene. The analysis was performed using the primers for amplifying the sucA gene by PCR (SEQ ID NOS: 5 and 6), and strains that were determined to contain a smaller PCR product than the PCR product using chromosomal DNA of the ATCC13869 strain, as a template. In this way, sucA-disrupted strains were selected and used in the following experiment.

[0100] The L-glutamic acid-producing ability of the sucA-disrupted strains were evaluated by the following method. The strains were cultured on CM-Dex plate medium, and each of the sucA gene-disrupted strains was seeded in a Sakaguchi flask containing 20 ml of a medium containing 30 g of glucose, 1 g of KH_2PO_4, 0.4 g of $MgSO_4$, 15 g of $(NH_4)_2SO_4$, 0.01 g of $FeSO_4 \cdot 7H_2O$, 0.01 g of $MnSO_4 \cdot 7H_2O$, 13.7 ml of soybean hydrolysate, 200 μg of thiamine chloride, 300 μg of biotin, and 50 g of $CaCO_3$ in 1 L of pure water (pH was adjusted to 8.0 with KOH), followed by a shaking culture at 31.5°C. After the culture, the amount of L-glutamic acid which had accumulated in the medium was determined, and the strain that exhibited the largest L-glutamic acid production yield was selected and named ATCC13869ΔsucA.

(1-2) Introduction of a mutant yggB gene

[0101] A mutant yggB gene prepared by inserting the IS (Insertion sequence) into the C-terminus of the yggB gene was introduced into the ATCC13869ΔsucA strain (Fig. 1). The nucleotide sequence of the mutant yggB gene and the amino acid sequence encoded thereby are shown in SEQ ID NOS: 7 and 8, respectively. The mutant yggB gene-introduced strain was named ATCC13869ΔsucAyggB::IS.
Alternatively, the strain in which the IS is inserted into the C-terminal region of the yggB gene can also be constructed by amplifying three fragments with PCR as follows.
First, PCR is performed using the oligonucleotides of SEQ ID NOS: 44 and 45 as primers and the chromosomal DNA from the ATCC13869 strain as a template, to thereby obtain an upstream fragment of the yggB gene. Another PCR is performed using the oligonucleotides of SEQ ID NOS: 46 and 47 as primers and the chromosomal DNA from the ATCC13869 strain as a template, to thereby obtain the IS fragment. The oligonucleotides of SEQ ID NOS: 45 and 46 are partially complementary to each other. Next, PCR is performed using approximately equal molar amounts of these PCR products as templates and the oligonucleotides of SEQ ID NOS: 44 and 47 as primers, to thereby prepare a fragment where the yggB fragment is inserted upstream of the IS.
PCR is performed using the oligonucleotides of SEQ ID NOS: 48 and 49 as primers and the chromosomal DNA from the ATCC 13 869 strain as a template, to thereby obtain a downstream fragment of the yggB gene. The oligonucleotides of SEQ ID NOS: 47 and 48 are complementary to each other. The yggB downstream fragment and the fragment described above comprising the yggB fragment inserted upstream of the IS are mixed in approximately equal molar amounts, and PCR is performed using the oligonucleotides of SEQ ID NOS: 50 and 51 as primers, to thereby obtain a yggB fragment into which the IS is inserted (yggB::IS). The fragment is treated with SacI and inserted into the SacI site of pBS4S (WO 2005/113745 and 2005/113744), to thereby construct a plasmid which can be used to replace the yggB gene on a chromosome with the yggB::IS. The ATCC13869 strain has a plurality of similar IS sequences. Therefore, to obtain an IS-inserted yggB gene completely corresponding to SEQ ID NO: 7, it is necessary to confirm the nucleotide sequence of the plasmid to determine whether the plasmid has the same sequence. However, differences in some nucleotides in an IS-derived region does not have a great influence on the function of the IS-inserted yggB gene. The resultant plasmid is used to replace the yggB gene on the chromosome by a conventional method, resulting in a strain in which the IS is inserted into the chromosomal yggB gene.

(1-3) Construction of a pta-amplified strain

[0102] To construct a strain in which the expression of a phosphotransacetylase (pta) gene is enhanced, a phosphotransacetylase (pta) gene derived from *C. glutamicum* ATCC 13 869 strain was amplified by PCR and cloned into the shuttle vector pVC7. pVC7 (US 20030134397) is constructed by inserting a fragment obtained by digesting pAM330 (a cryptic plasmid from the ATCC13869 strain (GenBank Database Accession No.D00038)) with HindIII and blunt-ending, into the BsaAI site of pHSG399 (Takara Bio). PCR was performed using synthetic DNAs (SEQ ID NOS: 9 and 10) designed based on the known nucleotide sequence of the phosphotransacetylase gene of *Corynebacterium glutamicum* ATCC 13032 (GenBank Database Accession No.NC_003450, SEQ ID NO: 40), to thereby obtain a gene fragment containing the entire ORF of the phosphotransacetylase gene and its promoter region. This PCR product was purified by a conventional method and digested with BamH I and Kpn I, and inserted between the BamH I site and Kpn I site in pVC7. This plasmid was used to transform competent cells of *Escherichia coli* DH5α (Takara Bio Inc.), and the transformed cells were cultured overnight on an LB plate containing 100 μM of IPTG, 40 μg/ml of X-Gal, and 25 μg/ml of chloramphenicol (Cm). Thereafter, single colonies of transformants were isolated by selecting the white colonies which appeared. Plasmids were extracted from these transformants, and the plasmid containing the objective PCR product was selected and named pVC7-pta.

(1-4) Confirmation of enhanced PTA activity in the ATCC13869ΔsucAyggB::IS strain

[0103] ATCC13869ΔsucAyggB::IS strain was transformed with pVK9. The transformation was performed by the electric pulse method, and the cells were cultured at 31.5°C for about 30 hours on a CM-Dex plate containing 25 μg/ml of kanamycin. This strain containing the pVK9 was named ATCC13869ΔsucAyggB::IS(pVK9).
pVK9 (US Patent 20050196846) is a shuttle vector. It is constructed by inserting a fragment obtained by digesting pHK4 (JP-A-05-007491) with BamHI and KpnI and blunt-ending (a fragment able to automatically replicate in coryneform bacteria), into blunt-ended AvaII site of pHSG299 (TAKARA BIO INC.).
Subsequently, ATCC13869ΔsucAyggB::IS(pVK9) strain was transformed separately with pVC7 (a control plasmid) and pVC7-pta (a plasmid for amplifying PTA). The transformation was performed by the electric pulse method, and the cells were cultured 31.5°C for about 30 hours on a CM-Dex plate containing 25 μg/ml of kanamycin and 5 μg/ml of chloramphenicol. The strains into which the above-described plasmids were introduced were named ATCC13869ΔsucAyggB::

IS (pVK9, pVC7) and
ATCC13869ΔsucAyggB::IS(pVK9, pVC7-pta), respectively.
To determine the PTA activity of these strains, a crude enzyme solution was prepared by the following method. First, each strain was cultured in CM-Dex liquid medium at 31.5°C. The OD 660 was determined using Taitec Mini photo 518R digital colorimeter, and the culture was continued until the OD reached 0.6 to 0.9, followed by collection of the cells. The following operations were performed at 4°C. The cells were washed with 50 mM Tris/HCl (pH 7.0) solution twice and then suspended in a buffer (50 mM Tris/HCl (pH 7.0), 10 mM $MgCl_2$, 1 mM EDTA, 1 mM DTT, and 30% (w/v) glycerol) to 4 g (wet weight)/ml. The cells were homogenized with an ultrasonic homogenizer (Bioruptor) and centrifuged (15,000 g, 60 min). The supernatant was used as the crude enzyme solution. The procedures for quantifying the protein level in the crude enzyme solution are as follows. Each crude enzyme solution and BSA (bovine serum albumin) with a known concentration (for creating a calibration curve) was allowed to react with a CBB solution (Nacalai tesque protein assay CBB solution) to develop a color, and then the protein concentration was quantified by determining the OD 595 nm using an apparatus for determining enzymatic activities (Molecular Devices, spectra max 190).
Subsequently, the PTA activity was determined with reference to the known method (D. J. Reinscheid, S. Schnicke, D. Rittmann, U. Zahnow, H. Sahm and B. J. Eikmanns (1999) Micobiology 145: 503-513). Specific procedures are shown below. The enzymatic reaction was initiated by adding the crude enzyme solution to the reaction solution of 100 mM of Tris/HCl (pH 7.6), 5 mM of $MgCl_2$, 0.5 mM of L-cysteine hydrochloride, 20 mM of NH_4Cl_2, 1 mM of CoA, and 20 mM of acetyl phosphate. The generation of acetyl-CoA was detected by determining the OD 232 nm using an apparatus for determining enzymatic activities (Molecular Devices, spectra max 190), to thereby determine the PTA activity. Table 1 shows that ATCC 13869ΔsucAyggB::IS(pVK9, pVC7-pta) has a higher PTA activity' as compared to the control strain.
[0104]

[Table 1]

	ΔABS/sec/mg	Relative activity to control defined as 1.
ATCC13869ΔsucAyggB::IS(pVK9, pVC7)	0.365	1 (Control)
ATCC13869ΔsucAyggB::IS(pVK9, pVC7-pta)	1.550	4.27

Example 2: L-glutamic acid production by the PTA-enhanced ATCC13869AsucAyggB::IS strain

[0105] The effect of enhancing PTA activity on the yield of L-glutamic acid during fermentation was evaluated by culturing *C. glutamicum* ATCC13869ΔsucAyggB::IS(pVK9, pVC7) strain and ATCC13869ΔsucAyggB::IS(pVK9, pVC7-pta) strain by the same method as shown above (1-1). The results are shown in Table 2. The yield of L-glutamic acid by the PTA-enhanced strain was found to be higher than that of the control strain.
[0106]

[Table 2]

	Fermentation yield of L-glutamic acid (mean ± standard deviation % (n = the number of samples))
ATCC13869ΔsucAyggB::IS(pVK9, pVC7)	47.0 ± 0.2 (n=4)
ATCC13869ΔsucAyggB::IS(pVK9, pVC7-pta)	50.5 ± 0.9 (n=4)

Example 3: Production of L-glutamic acid by the strain in which activities of PTA and phosphoketolase are enhanced

[0107] Next, the combined effect of the enhanced phosphotransacetylase activity and enhanced phosphoketolase activity on L-glutamic acid production was examined.
A phosphoketolase expression plasmid was constructed as follows.

(A) Construction of pVK9-xfp

[0108] To amplify the phosphoketolase gene (the sequence information has been disclosed: AY518213:gi:41056820) by PCR and insert the gene into the shuttle vector pVK9 used in Example 1, the chromosomal DNA was extracted from *Bifidobacterium animalis* using Wizard Genomic Purification Kit (Promega Corporation). PCR was performed using a chromosomal DNA of *B. animalis* as a template and synthetic DNAs of SEQ ID NOS: 11 and 12 as primers, to thereby obtain a gene fragment containing the entire ORF of the phosphoketolase gene and its promoter region. This PCR

product was purified by a conventional method and then digested with Xba I, followed by insertion into the Xba I site of pVK9. Competent cells of *Escherichia coli* DH5α (Takara Bio Inc.) were transformed with this DNA, and the transformed cells were cultured overnight on an LB plate containing 100 μM of IPTG, 40 μg/ml of X-Gal, and 25 μg/ml of Km. Thereafter, single colonies of transformants were isolated by selecting the white colonies which appeared. Plasmids were extracted from the transformants, and those containing the desired PCR product were selected and named pVK9-xfp.

(B) Construction of pVK9-PS2_xfp

[0109] The overlap PCR method (R.M. Horton, H.D. Hunt, S.N. Ho, J.K. Pullen, and L.R. Pease (1989) Gene 77: 61-68) was performed to obtain a DNA fragment in which a promoter region of the phosphoketolase gene of *B. animalis* was replaced with the PS2 promoter. Specifically, PCR was performed using pPSTG1 (Y. Kikuchi, M. Date, K. Yokoyama, Y Umezawa, and H. Matsui (2003) Appl. Environ. Microbiol. 69: 358-366) as a template and synthetic DNAs of SEQ ID NOS: 13 and 14 as primers, to thereby obtain an amplified product of the PS2 promoter. Another PCR was performed using pVK9-xfp as a template and synthetic DNAs of SEQ ID NOS: 15 and 16 as primers, to thereby obtain an amplified product of the *B. animalis* phosphoketolase gene. The primers of SEQ ID NOS: 14 and 16 are complementary to each other. Next, to obtain a fragment where the PS2 promoter is inserted upstream of the *B. animalis* phosphoketolase gene, the PS2 promoter and *B. animalis* phosphoketolase gene product were mixed in approximately equal amounts, and PCR was performed using the mixture as templates and synthetic DNAs of SEQ ID NOS: 12 and 17 as primers. This PCR product was purified by a conventional method and then digested with Xba I, followed by insertion into the Xba I site of pVK9. The DNA was used to transform competent cells of *Escherichia coli* DH5α (Takara Bio Inc.), and the transformed cells were cultured overnight on an LB plate containing 100 μM of IPTG, 40 μg/ml of X-Gal, and 25 μg/ml of Km. Thereafter, single colonies of transformants were isolated by selecting the white colonies which appeared. Plasmids were extracted from these transformants, and those containing the plasmid having the desired PCR product were selected and named pVK9-PS2_xfp.

(C) Production of L-glutamic acid using a strain with enhanced activities of PTA and phosphoketolase

[0110] *C. glutamicum* ATCC13869ΔsucAyggB::IS strain was transformed with pVK9_PS2_xfp in accordance with the above-described method, to thereby obtain a transformant strain, which was named ATCC13869ΔsucAyggB::IS (pVK9_PS2_xfp). Next, the ATCC13869ΔsucAyggB::IS(pVK9_PS2_xfp) strain was transformed separately with pVC7 (a plasmid for control) and pVC7-pta (a plasmid for amplifying PTA) and the transformed strains were named ATCC13869ΔsucAyggB::IS(pVK9_PS2_xfp, pVC7) and ATCC13869ΔsucAyggB::IS(pVK9_PS2_xfp, pVC7-pta), respectively.
C. glutamicum ATCC13869ΔsucAyggB::IS(pVK9, pVC7), ATCC13869ΔsucAyggB::IS(pVK9, pVC7-pta), ATCC13869ΔsucAyggB::IS(pVK9_PS2_xfp, pVC7), and ATCC13869ΔsucAyggB::IS(pVK9_PS2_xfp, pVC7-pta) strains were cultured in accordance with the method of (1-1) of Example 1. The results are shown in Table 3. The yield of L-glutamic acid from the strain having both enhanced PTA and phosphoketolase activity was found to be higher as compared to the strains in which either of the PTA or phosphoketolase was enhanced.

[0111]

[Table 3]

	Fermentation yield of L-glutamic acid (%) (mean ± standard deviation (n = the number of samples))
ATCC13869ΔsucAyggB::IS(pVK9, pVC7)	47.0 ± 0.2 (n=4)
ATCC13869ΔsucAyggB::IS(pVK9, pVC7-pta)	50.5 ± 0.9 (n=4)
ATCC13869ΔsucAyggB::IS(pVK9 PS2_xfp, pVC7)	54.5 ± 0.2 (n=3)
ATCC13869ΔsucAyggB::IS(pVK9_PS2_xfp, pVC7-pta)	56.6 ± 0.7 (n=6)

Example 4: Construction of a strain in which PTA activity is enhanced by disrupting a ramB gene that encodes a transcription factor, and evaluation of its effect

[0112] It is known that the expression of the PTA gene is negatively regulated by the transcription factor RamB, and the PTA activity is increased in a ramB gene-deficient strain (Journal of Bacteriology May 2004 2798-2809). Therefore, a ramB-deficient strain is constructed, and its effect on the yield of L-glutaxnic acid during fermentation is examined.

(4-1) Cloning of a fragment for disrupting the ramB gene

[0113] A ramB gene is disrupted using pBS5T, which is a temperature-sensitive plasmid carrying the sacB gene (WO 2005/11374s and WO 2005/113744). A gene fragment which is derived from the ramB gene of the ATCC13869 strain and has a deletion in the ORF of the ramB gene is obtained by the overlap PCR method using synthetic DNAs designed based on the known nucleotide sequence of the gene from *Corynebacterium glutamicum* ATCC13032 (GenBank Database Accession No. NC_003450, the nucleotide sequence of ramB gene and the amino acid sequence encoded thereby are shown in SEQ ID NOS: 36 and 37, respectively) as primers. Specifically, PCR is performed by using the chromosomal DNA of *C. glutamicum* ATCC13869 strain as a template and synthetic DNAs of SEQ ID NOS: 19 and 20 as primers, to thereby obtain an amplified product corresponding to the N-terminal region of the ramB gene. Another PCR is performed to amplify a fragment corresponding to the C-terminal region of the ramB gene, using the chromosomal DNA of ATCC 13869 strain as a template and synthetic DNAs of SEQ ID NOS: 18 and 21 as primers. The DNAs of SEQ ID NOS: 18 and 19 are complementary to each other, and they are designed so that a mutant ramB gene which is missing the entire ORF is amplified. Subsequently, to obtain the mutant ramB gene fragment missing an internal sequence, the above-described amplified products corresponding to the N-terminal region and the C-terminal region of the ramB gene are mixed in approximately equal molar amounts, and PCR is performed using the mixture as templates and using synthetic DNAs of SEQ ID NOS: 22 and 23 as primers. This PCR product is purified by a conventional method, and then digested with Xba I, followed by insertion into the Xba I site of the above-described pBS5T. Competent cells of *Escherichia coli* DH5α (Takara Bio Inc.) are transformed with this DNA, and the cells are cultured overnight on LB medium containing 100 μM of IPTG, 40 μg/ml of X-Gal, and 25 μg/ml of Km. Thereafter, single colonies of transformants are isolated by selecting the white colonies which appear. Plasmids are extracted from these transformants, and those containing the desired PCR are selected and named pBS5T-ramB.

(4-2) Construction of a ramB gene-disrupted strain

[0114] First, ATCC13869ΔsucAyggB::IS strain is transformed by the electric pulse method with a high concentration of pBS5T-ramB, and the transformed cells are cultured at 25°C for about 60 hours on a CM-Dex plate containing 25 μg/ml of kanamycin. This transformant is cultured with shaking at 34°C overnight in CM-Dex liquid medium and after appropriate dilution, the cells are cultured at 34°C for about 30 hours in CM-Dex medium containing 25 μg/ml of kanamycin. In the strain which grew in this medium, the kanamycin-resistant gene and the sacB gene derived from the plasmid are integrated into the chromosome by homologous recombination between the mutant ramB gene of the plasmid and the ramB gene on the chromosome of the ATCC13869ΔsucAyggB::IS strain.
Next, these single-crossover recombinants are cultured at 31.5°C overnight in CM-Dex liquid medium containing no kanamycin, and after appropriate dilution, the cells are cultured at 34°C for about 30 hours in 10% sucrose-containing Dex-S10 medium (100 g/L sucrose, 10 g/L polypeptone, 10 g/L yeast extract, 1 g/L KH_2PO_4, 0.4 g/L $MgSO_4 \cdot 7H_2O$, 0.01 g/L $FeSO_4 \cdot 7H_2O$, 0.01 g/L $MnSO_4 \cdot 4H_2O$, 3 g/L urea, 1.2 g/L soybean hydrolysate, 10 μg/L biotin, 2 g/l sodium acetate, pH 7.5 (KOH)) with no kanamycin. As a result, strains which are sucrose-insensitive due to the curing of the sacB gene by the second homologous recombination are obtained.
The strains thus obtained include one in which the ramB gene is replaced by the mutant gene derived from pBS5T-ramB, and one in which the ramB gene reverts to the wild-type gene. Whether the ramB gene is the mutant gene or the wild-type gene can be confirmed by directly subjecting bacterial cells obtained by culture on Dex-S10 agar medium to PCR. A strain having only the mutant ramB gene is selected and named ATCC3869ΔsucAyggB::ISΔramB.

(4-3) L-glutamic acid production using the ramB gene-disrupted strain

[0115] ATCC13869Δsuc.AyggB::ISΔramB is transformed with pVK9 (a plasmid for control) and pVK9_PS2_xfp (a plasmid for amplifying the phosphoketolase gene (PKT) separately. The transformation is performed by the electric pulse method, and the cells are cultured at 31.5°C for about 30 hours on a CM-Dex plate containing 25 μg/ml of kanamycin. The plasmid-introduced strains are named ATCC13869ΔsucAyggB::ISΔramB(pVK9) and ATCC13869ΔsucAyggB::ISΔramB(pVK9_PS2_xfp), respectively. These strains are evaluated by culture in accordance with the method of (1-1) of Example 1 together with ATCC13869ΔsucAyggB::IS(pVK9) strain andATCC13869ΔsucAyggB::IS(pVK9_PS2_xfp) strain constructed in Example 1. This procedure confirms the improvement of the L-glutamic acid yield by ramB gene disruption and by a combination of ramB gene disruption and PKT enhancement.

Example 5: Construction of a PTA activity-enhanced strain by modification of a promoter of the pta gene and evaluation of the effect

[0116] The PTA activity can be enhanced by modifying a promoter of the pta gene. It is known that a the pta gene

promoter has two regions that are thought to be involved in the RamB-mediated negative regulation that affects the PTA activity (J Bacteriol. 2004 May; 186(9): 2798-2809.). Therefore, strains which are modified in only one of the RamB-binding sites in the pta promoter or both of the RamB-binding sites in the pta promoter, are constructed to examine the effect on the L-glutamic acid fermentation yield.

(5-1) Construction of plasmids mutating the pta promoter, pBS5T-m1PTA, pBS5T-m2PTA, PBS5T-mlm2PTA

[0117] A gene fragment containing the ORF region and the promoter region of the pta gene derived from ATCC13869 is obtained by PCR using synthetic DNAs designed based on the known nucleotide sequence of the gene from *Corynebacteriurn glutamicum* ATCC13032 (GenBank Database Accession No.NC_003450). Specifically, for amplifying the promoter region of the pta gene, PCR is performed using the chromosomal DNA of C. *glutamicum* ATCC13869 strain as a template and synthetic DNAs of SEQ ID NOS: 24 and 25 as primers. This PCR product is purified by a conventional method and digested with Xba I, followed by insertion into the Xba I region of pUC19 (Takara Bio Inc.). Competent cells of *Escherichia coli* DH5α (Takara Bio Inc.) are transformed with the DNA, and cultured overnight in LB medium containing 100 μM of IPTG, 40 μg/ml of X-Gal, and 50 μg/ml of Amp. Thereafter, single colonies of transformants are isolated by selecting the white colonies which appear. Plasmids are extracted from these transformants, and those containing the desired PCR product is selected and named pUC 19-PTA.
Next, a mutation is introduced into the pta promoter region using Site-Directed Mutagenesis Kits (STARATAGENE). PCR is performed using pUC 19-PTA as a template and synthetic DNAs of SEQ ID NOS: 26 and 27 as primers in accordance with the manual supplied with the kits, which results in a plasmid having a mutation in one of the first RamB-binding sites. This plasmid is named pUC19-m1PTA.
Next, PCR is performed using pUC 19-ml PTA as a template and synthetic DNAs of SEQ ID NOS: 24 and 25 as primers, to thereby obtain an amplified product containing the promoter region of the pta gene having the mutation. This PCR product is purified by a conventional method and inserted into the above-described SmaI region of pBS5T. Competent cells of *Escherichia coli* DH5α (Takara Bio Inc.) are transformed with this DNA, and the cells are cultured overnight in LB medium containing 100 μM of IPTG, 40 μg/ml of X-Gal, and 25 μg/ml of Km. Thereafter, single colonies of transformants are isolated by selecting the white colonies which appear. Plasmids are extracted from these transformants, and sequence analysis confirms that the plasmid has the desired mutation with no sequence errors in other nucleotides. Thus, the plasmid is named pB5ST-m1PTA.
In the same way, a plasmid having a mutation in the second RamB-binding site is constructed by performing PCR using synthetic DNAs of SEQ ID NOS: 28 and 29, and then inserting the desired sequence into pBS5T. This plasmid is named pBS5T-m2PTA. In addition, a plasmid having mutations in both RamB-binding sites is constructed by the same procedures as described above and named pBS5T-m1m2PTA.

(5-2) Construction of strains harboring the promoter-modified pta gene

[0118] Strains having modified PTA promoters are obtained from ATCC13869ΔsucAyggB::IS strain in accordance with the method of Example 4. The mutation is confirmed by analyzing the sequence of the promoter region. The strains with modified RamB-binding sites in the pta promoter are named ATCC 13869ΔsucAyggB::ISm1, ATCC13869ΔsucAyggB::ISm2, and ATCC13869ΔsucAyggB::ISm1m2.

(5-3) L-glutamic acid production by using the strains harboring the promoter-modified pta gene

[0119] The ATCC13869ΔsucAyggB::ISm1 strain, ATCC13869ΔsucAyggB::ISm2 strain, andATCC13869ΔsucAyggB::ISm1m2 strain are each transformed with pVK9 (a control plasmid) and pVK9_PS2_xfp (a plasmid for amplifying PKT). The transformation is performed by the electric pulse method, and the transformed cells are cultured at 31.5°C for about 30 hours on a CM-Dex plate containing 25 μg/ml of kanamycin. These strains are named ATCC13869ΔsucAyggB::ISm1 (pVK9), ATCC13869ΔsucAyggB::ISm1(pVK9_PS2_xfp), ATCC13869ΔsucAyggB::ISm2(pVK9), ATCC13869ΔsucAyggB::ISm2(pVK9_PS2_xfp), ATCC13869ΔsucAyggB::ISm1m2(pVK9), and ATCC13869ΔsucAm1m2(pVK9_PS2_xfp), and are evaluated by culture in accordance with the method of Example 1 together with ATCC13869ΔsucAyggB::IS(pVK9) and ATCC13869ΔsucA(pVK9_PS2_xfp), constructed in Example 1. Improvement of the L-glutamic acid yield by the pta promoter modification and the combination of the PTA promoter modification and PKT enhancement is confirmed.

Example 6

Construction of a strain in which PTA enhancement is combined with the enhancement of enzymes of anaplerotic pathway

[0120] In order to evaluate the effect of combined enhancement of PTA and phosphoenolpyruvate carboxylase (PPC) or pyruvate carboxylase, (PC), strains with enhanced activities of PTA and PPC or enhanced activities of PTA and PC are constructed.

(6-1) Construction of the ppc-expressing plasmid pVC-PPC

[0121] The ppc-expressing plasmid is constructed as follows. The ppc gene is amplified by performing PCR using synthetic oligonucleotides of SEQ ID NOS: 58 and 59 as primers and the chromosomal DNA of *C. glutamicum* ATCC 14067 strain as a template. The amplified DNA is blunt-ended and inserted into the SmaI site of pHSG399 (TAKARA BIO INC.), to thereby obtain a plasmid pPCF. Then, in order to remove the region upstream of the coding region of the ppc gene, pPCF is digested with DraI and SalI, and the obtained fragment is inserted into the SmaI site of pHSG398 (TAKARA BIO INC.) to obtain a plasmid having the ppc gene inserted in the reverse direction with respect to the lacZ gene, and is named pPCFds. Then, a DNA fragment containing an aspartokinase promoter and a region able to autonomously replicate in coryneform bacterium is prepared by digesting p399AKYB (JP6-62866) with PstI and ApaI and then blunt-ending, and the obtained fragment is inserted into the pPCFds which has been digested with SalI and blunt-ended, to ligate the aspartokinase promoter upstream of the ppc gene. A plasmid in which the aspartokinase promoter and the ppc gene are linked in the same direction was selected and named pAKPFds. The region in pAKPFds involved in autonomous replication in coryneform bacterium is derived from pHM1519, since the autonomous replication region of pVK9_PS2_xfp, and therefore, pAKPFds cannot co-exist with pVK9_PS2_xfp in coryneform bacterium. So, PCR is performing by using synthetic oligonucleotides of SEQ ID NOS: 60 and 61 as primers and pAKPFds as a template, and the amplified fragment is digested with KpnI and inserted into the KpnI site of pVC7 (JP2000-201692), which contains an autonomous replication region from pAM330. The obtained plasmid was named pVC-PPC.

(6-2) Construction of a strain in which the pta gene and the ppc gene are amplified, and a strain in which the pta gene and the pc gene are amplified

[0122] pVC7 (control), pVC-PPC, or pBPYC6 (pc gene-expressing plasmid: JP2000-201692) is used to transform ATCC13869ΔsucAyggB::IS(pVK9) (see Example 2), ATCC13869ΔsucA(pVK9_PS2_xfp) (see Example 3), and ATCC13869ΔsucAyggB::IS m1(pVK9), ATCC13869ΔsucAyggB::IS m1(pVK9_PS2_xfp), ATCC13869ΔsucAyggB::IS m2(pVK9), ATCC13869ΔsucAyggB::IS m2(pVK9_PS2_xfp), ATCC13869ΔsucAyggB::IS m1m2(pVK9), and ATCC13869ΔsucAyggB::IS m1m2(pVK9_PS2_xfp) (see Example 5). These transformants are cultured and L-glutamic acid production is evaluated according to the method of (1-1) of Example 1. Thereby, improvement of L-glutamic acid production is confirmed by combining the modification of the pta promoter with the enhancement of PC or PPC, or by combining the modification of the pta promoter with the enhancement of PKT and PC or PPC.

INDUSTRIAL APPLICABILITY

[0123] The production method of the present invention enables efficient production of L-amino acids such as L-glutamic acid, L-glutamine, L-proline and L-arginine.

SEQUENCE LISTING

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<120> Method for producing L-amino acid

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tcggcgcagc ccctcagtac ctgttttggc tgctcgcgct ccctgtcatc ttcggttact   4715
gggttctaaa atcatccacg atcgttgatg aacagggcat caccgcaaac tacgccttca   4775
agggcaaaaa ggttgtggcc tgggaagacc tcgcaggaat cggattcaag ggtgcccgca   4835
ctttcgctcg caccacctcc gatgcagaag tcaccctccc cggcgtcacc ttcaactccc   4895
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<211> 423
<212> PRT
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<400> 8

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1               5                  10                  15
Trp Ile Val Asp Thr Gly Phe Asp Val Ala Ile Ile Leu Val Leu Ala
            20                  25                  30
Phe Leu Ile Pro Arg Ile Gly Arg Leu Ala Met Arg Ile Ile Lys Gln
        35                  40                  45
Arg Val Glu Ser Ala Ala Asp Ala Asp Thr Thr Lys Asn Gln Leu Ala
    50                  55                  60
Phe Ala Gly Val Gly Val Tyr Ile Ala Gln Ile Val Ala Phe Phe Met
65                  70                  75                  80
Leu Ala Val Ser Ala Met Gln Ala Phe Gly Phe Ser Leu Ala Gly Ala
                85                  90                  95
Ala Ile Pro Ala Thr Ile Ala Ser Ala Ala Ile Gly Leu Gly Ala Gln
            100                 105                 110
Ser Ile Val Ala Asp Phe Leu Ala Gly Phe Phe Ile Leu Thr Glu Lys
        115                 120                 125
Gln Phe Gly Val Gly Asp Trp Val Arg Phe Glu Gly Asn Gly Ile Val
    130                 135                 140
Val Glu Gly Thr Val Ile Glu Ile Thr Met Arg Ala Thr Lys Ile Arg
145                 150                 155                 160
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Thr Ile Ala Gln Glu Thr Val Ile Ile Pro Asn Ser Thr Ala Lys Val
                165                 170                 175
Cys Ile Asn Asn Ser Asn Asn Trp Ser Arg Ala Val Val Val Ile Pro
            180                 185                 190
Ile Pro Met Leu Gly Ser Glu Asn Ile Thr Asp Val Ile Ala Arg Ser
        195                 200                 205
Glu Ala Ala Thr Arg Arg Ala Leu Gly Gln Glu Lys Ile Ala Pro Glu
    210                 215                 220
Ile Leu Gly Glu Leu Asp Val His Pro Ala Thr Glu Val Thr Pro Pro
225                 230                 235                 240
Thr Val Val Gly Met Pro Trp Met Val Thr Met Arg Phe Leu Val Gln
                245                 250                 255
Val Thr Ala Gly Asn Gln Trp Leu Val Glu Arg Ala Ile Arg Thr Glu
            260                 265                 270
Ile Ile Asn Glu Phe Trp Glu Glu Tyr Gly Ser Ala Thr Thr Thr Ser
        275                 280                 285
Gly Thr Leu Ile Asp Ser Leu His Val Glu His Glu Glu Pro Lys Thr
    290                 295                 300
Ser Leu Ile Asp Ala Ser Pro Gln Ala Leu Lys Glu Pro Lys Pro Glu
305                 310                 315                 320
Ala Ala Ala Thr Val Ala Ser Leu Ala Ala Ser Ser Asn Asp Asp Ala
                325                 330                 335
Asp Asn Ala Asp Ala Ser Ala Ile Asn Ala Gly Asn Pro Glu Lys Glu
            340                 345                 350
Leu Asp Ser Asp Val Leu Glu Gln Glu Leu Ser Ser Glu Glu Pro Glu
        355                 360                 365
Glu Thr Ala Lys Pro Asp His Ser Leu Arg Gly Phe Phe Arg Thr Asp
    370                 375                 380
Tyr Tyr Pro Asn Arg Trp Gln Lys Ile Leu Ser Phe Gly Gly Arg Val
385                 390                 395                 400
Arg Met Ser Thr Ser Leu Leu Leu Gly Ala Leu Leu Leu Leu Ser Leu
                405                 410                 415
Phe Lys Gly Leu Phe Leu Phe
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cggggtacct ataaagttcg attccttaaa ggggttc 37

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tggagcaaca cccatggtca ggtcgaggat 30

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<210> 27
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<400> 27
gttgcgaagc acccttgcc tcgtttttag cggtacttta c 41

<210> 28
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cgtccgcctt gcgtacagcc cggctcgcct cgttttattt tgtaattttc 50

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<220>
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gcgccaactg atcgcgctgg cgcgcgccga actcatcgag ccttccatca tgcttctcga    180
cgaagccacc tccaccctcg accccgccac cgaagccgtt atcctcaacg cctccgatcg    240
agtcactaag ggacgcacca gcatcatcgt cgcgcaccgc ttggcaaccg ctaaaagggc    300
cgaccgtatt cttgttgttg aacaaggacg tatcattgag gacggatctc acgacgcgtt    360
gttgtctgct aacggcacct acgcccgcat gtggcattta atggcctgac acgttatttt    420
taggagaact gtcaacaaat ta atg cta caa ctg ggg ctt agg cat aat cag      472
                         Met Leu Gln Leu Gly Leu Arg His Asn Gln
                         1               5                  10
cca acg acc aac gtt aca gtg gat aaa ata aag ctc aat aaa ccc tca      520
Pro Thr Thr Asn Val Thr Val Asp Lys Ile Lys Leu Asn Lys Pro Ser
                    15                  20                  25
aga agc aag gaa aag agg cga gta cct gcc gtg agc agc gct agt act      568
Arg Ser Lys Glu Lys Arg Arg Val Pro Ala Val Ser Ser Ala Ser Thr
                30                  35                  40
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ttc ggc cag aat gcg tgg ctg gta gac gag atg ttc cag cag ttc cag      616
Phe Gly Gln Asn Ala Trp Leu Val Asp Glu Met Phe Gln Gln Phe Gln
        45                  50                  55
aag gac ccc aag tcc gtg gac aag gaa tgg aga gaa ctc ttt gag gcg      664
Lys Asp Pro Lys Ser Val Asp Lys Glu Trp Arg Glu Leu Phe Glu Ala
    60                  65                  70
cag ggg gga cca aat gct acc ccc gct aca aca gaa gca cag cct tca      712
Gln Gly Gly Pro Asn Ala Thr Pro Ala Thr Thr Glu Ala Gln Pro Ser
75                  80                  85                  90
gcg ccc aag gag tct gcg aaa cca gca cca aag gct gcc cct gca gcc      760
Ala Pro Lys Glu Ser Ala Lys Pro Ala Pro Lys Ala Ala Pro Ala Ala
                95                  100                 105
aag gca gca ccg cgc gta gaa acc aag ccg gcc gcc aag acc gcc cct      808
Lys Ala Ala Pro Arg Val Glu Thr Lys Pro Ala Ala Lys Thr Ala Pro
            110                 115                 120
aag gcc aag gag tcc tca gtg cca cag caa cct aag ctt ccg gag cca      856
Lys Ala Lys Glu Ser Ser Val Pro Gln Gln Pro Lys Leu Pro Glu Pro
        125                 130                 135
gga caa acc cca atc agg ggt att ttc aag tcc atc gcg aag aac atg      904
Gly Gln Thr Pro Ile Arg Gly Ile Phe Lys Ser Ile Ala Lys Asn Met
    140                 145                 150
gat atc tcc ctg gaa atc cca acc gca acc tcg gtt cgc gat atg cca      952
Asp Ile Ser Leu Glu Ile Pro Thr Ala Thr Ser Val Arg Asp Met Pro
155                 160                 165                 170
gct cgc ctc atg ttc gaa aac cgc gcg atg gtc aac gat cag ctc aag     1000
Ala Arg Leu Met Phe Glu Asn Arg Ala Met Val Asn Asp Gln Leu Lys
                175                 180                 185
cgc acc cgc ggt ggc aag atc tcc ttc acc cac atc att ggc tac gcc     1048
Arg Thr Arg Gly Gly Lys Ile Ser Phe Thr His Ile Ile Gly Tyr Ala
            190                 195                 200
atg gtg aag gca gtc atg gct cac ccg gac atg aac aac tcc tac gac     1096
Met Val Lys Ala Val Met Ala His Pro Asp Met Asn Asn Ser Tyr Asp
        205                 210                 215
gtc atc gac ggc aag cca acc ctg atc gtg cct gag cac atc aac ctg     1144
Val Ile Asp Gly Lys Pro Thr Leu Ile Val Pro Glu His Ile Asn Leu
    220                 225                 230
ggc ctt gcc atc gac ctt cct cag aag gac ggc tcc cgc gca ctt gtc     1192
Gly Leu Ala Ile Asp Leu Pro Gln Lys Asp Gly Ser Arg Ala Leu Val
235                 240                 245                 250
gta gca gcc atc aag gaa acc gag aag atg aac ttc tcc gag ttc ctc     1240
Val Ala Ala Ile Lys Glu Thr Glu Lys Met Asn Phe Ser Glu Phe Leu
                255                 260                 265
gca gca tac gaa gac atc gtg aca cgc tcc cgc aag ggc aag ctc acc     1288
Ala Ala Tyr Glu Asp Ile Val Thr Arg Ser Arg Lys Gly Lys Leu Thr
            270                 275                 280
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atg gat gac tac cag ggc gtt acc gtt tcc ttg acc aac cca ggt ggc    1336
Met Asp Asp Tyr Gln Gly Val Thr Val Ser Leu Thr Asn Pro Gly Gly
        285                 290                 295
atc ggt acc cgc cac tct gtc cca cgt ctg acc aag ggc cag ggc acc    1384
Ile Gly Thr Arg His Ser Val Pro Arg Leu Thr Lys Gly Gln Gly Thr
    300                 305                 310
atc atc ggt gtc ggt tcc atg gat tac cca gca gag ttc cag ggc gct    1432
Ile Ile Gly Val Gly Ser Met Asp Tyr Pro Ala Glu Phe Gln Gly Ala
315                 320                 325                 330
tcc gaa gac cgc ctt gca gag ctc ggc gtt gga aag ctt gtc acc atc    1480
Ser Glu Asp Arg Leu Ala Glu Leu Gly Val Gly Lys Leu Val Thr Ile
                335                 340                 345
acc tcc acc tac gat cac cgc gtg atc cag ggt gct gtg tcc ggt gaa    1528
Thr Ser Thr Tyr Asp His Arg Val Ile Gln Gly Ala Val Ser Gly Glu
            350                 355                 360
ttc ctg cgt acc atg tct cgc ctg ctc acc gat gat tcc ttc tgg gat    1576
Phe Leu Arg Thr Met Ser Arg Leu Leu Thr Asp Asp Ser Phe Trp Asp
        365                 370                 375
gag atc ttc gac gca atg aac gtt cct tac acc cca atg cgt tgg gca    1624
Glu Ile Phe Asp Ala Met Asn Val Pro Tyr Thr Pro Met Arg Trp Ala
    380                 385                 390
cag gac gtt cca aac acc ggt gtt gat aag aac acc cgc gtc atg cag    1672
Gln Asp Val Pro Asn Thr Gly Val Asp Lys Asn Thr Arg Val Met Gln
395                 400                 405                 410
ctc att gag gca tac cgc tcc cgt gga cac ctc atc gct gac acc aac    1720
Leu Ile Glu Ala Tyr Arg Ser Arg Gly His Leu Ile Ala Asp Thr Asn
                415                 420                 425
cca ctt tca tgg gtt cag cct ggc atg cca gtt cca gac cac cgc gac    1768
Pro Leu Ser Trp Val Gln Pro Gly Met Pro Val Pro Asp His Arg Asp
            430                 435                 440
ctc gac atc gag acc cac agc ctg acc atc tgg gat ctg gac cgt acc    1816
Leu Asp Ile Glu Thr His Ser Leu Thr Ile Trp Asp Leu Asp Arg Thr
        445                 450                 455
ttc agc gtc ggt ggc ttc ggc ggc aag gag acc atg acc ctg cgc gag    1864
Phe Ser Val Gly Gly Phe Gly Gly Lys Glu Thr Met Thr Leu Arg Glu
    460                 465                 470
gta ctg tcc cgc ctg cgc gct gcc tac acc ttg aag gtc ggc tcc gaa    1912
Val Leu Ser Arg Leu Arg Ala Ala Tyr Thr Leu Lys Val Gly Ser Glu
475                 480                 485                 490
tac acc cac atc ctg gac cgc gac gag cgc acc tgg ctg cag gac cgc    1960
Tyr Thr His Ile Leu Asp Arg Asp Glu Arg Thr Trp Leu Gln Asp Arg
                495                 500                 505
ctc gaa gcc gga atg cca aag cca acc cag gca gag cag aag tac atc    2008
Leu Glu Ala Gly Met Pro Lys Pro Thr Gln Ala Glu Gln Lys Tyr Ile
            510                 515                 520
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ctg cag aag ctg aac gcc gca gag gct ttc gag aac ttc ctg cag acc      2056
Leu Gln Lys Leu Asn Ala Ala Glu Ala Phe Glu Asn Phe Leu Gln Thr
        525                 530                 535

aag tac gtc ggc cag aag cgc ttc tcc ctc gaa ggt gca gaa gct ctc      2104
Lys Tyr Val Gly Gln Lys Arg Phe Ser Leu Glu Gly Ala Glu Ala Leu
    540                 545                 550

atc cca ctg atg gac tcc gcc atc gac acc gcc gca ggc cag ggc ctc      2152
Ile Pro Leu Met Asp Ser Ala Ile Asp Thr Ala Ala Gly Gln Gly Leu
555                 560                 565                 570

gac gaa gtt gtc atc ggt atg cca cac cgt ggt cgc ctc aac gtg ctg      2200
Asp Glu Val Val Ile Gly Met Pro His Arg Gly Arg Leu Asn Val Leu
                575                 580                 585

ttc aac atc gtg ggc aag cca ctg gca tcc atc ttc aac gag ttt gaa      2248
Phe Asn Ile Val Gly Lys Pro Leu Ala Ser Ile Phe Asn Glu Phe Glu
            590                 595                 600

ggc caa atg gag cag ggc cag atc ggt ggc tcc ggt gac gtg aag tac      2296
Gly Gln Met Glu Gln Gly Gln Ile Gly Gly Ser Gly Asp Val Lys Tyr
        605                 610                 615

cac ctc ggt tcc gaa ggc cag cac ctg cag atg ttc ggc gac ggc gag      2344
His Leu Gly Ser Glu Gly Gln His Leu Gln Met Phe Gly Asp Gly Glu
    620                 625                 630

atc aag gtc tcc ctg act gct aac ccg tcc cac ctg gaa gct gtt aac      2392
Ile Lys Val Ser Leu Thr Ala Asn Pro Ser His Leu Glu Ala Val Asn
635                 640                 645                 650

cca gtg atg gaa ggt atc gtc cgc gca aag cag gac tac ctg gac aag      2440
Pro Val Met Glu Gly Ile Val Arg Ala Lys Gln Asp Tyr Leu Asp Lys
                655                 660                 665

ggc gta gac ggc aag act gtt gtg cca ctg ctg ctc cac ggt gac gct      2488
Gly Val Asp Gly Lys Thr Val Val Pro Leu Leu Leu His Gly Asp Ala
            670                 675                 680

gca ttc gca ggc ctg ggc atc gtg cca gaa acc atc aac ctg gct aag      2536
Ala Phe Ala Gly Leu Gly Ile Val Pro Glu Thr Ile Asn Leu Ala Lys
        685                 690                 695

ctg cgt ggc tac gac gtc gga ggc acc atc cac atc gtg gtg aac aac      2584
Leu Arg Gly Tyr Asp Val Gly Gly Thr Ile His Ile Val Val Asn Asn
    700                 705                 710

cag atc ggc ttc acc acc acc cca gac tcc agc cgc tcc atg cac tac      2632
Gln Ile Gly Phe Thr Thr Thr Pro Asp Ser Ser Arg Ser Met His Tyr
715                 720                 725                 730

gca acc gac tac gcc aag gca ttc ggc tgc cca gtc ttc cac gtc aat      2680
Ala Thr Asp Tyr Ala Lys Ala Phe Gly Cys Pro Val Phe His Val Asn
                735                 740                 745

ggt gat gac cca gag gca gtt gtc tgg gtt ggc cag ctg gca acc gag      2728
Gly Asp Asp Pro Glu Ala Val Val Trp Val Gly Gln Leu Ala Thr Glu
            750                 755                 760
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tac cgt cgt cgc ttc ggc aag gac gtc ttc atc gac ctc gtt tgc tac     2776
Tyr Arg Arg Arg Phe Gly Lys Asp Val Phe Ile Asp Leu Val Cys Tyr
        765                 770                 775

cgc ctc cgc ggc cac aac gaa gct gat gat cct tcc atg acc cag cca     2824
Arg Leu Arg Gly His Asn Glu Ala Asp Asp Pro Ser Met Thr Gln Pro
    780                 785                 790

aag atg tat gag ctc atc acc ggc cgc gag acc gtt cgt gct cag tac     2872
Lys Met Tyr Glu Leu Ile Thr Gly Arg Glu Thr Val Arg Ala Gln Tyr
795                 800                 805                 810

acc gaa gac ctg ctc gga cgt gga gac ctc tcc aac gaa gat gca gaa     2920
Thr Glu Asp Leu Leu Gly Arg Gly Asp Leu Ser Asn Glu Asp Ala Glu
                815                 820                 825

gca gtc gtc cgc gac ttc cac gac cag atg gaa tct gtg ttc aac gaa     2968
Ala Val Val Arg Asp Phe His Asp Gln Met Glu Ser Val Phe Asn Glu
            830                 835                 840

gtc aag gaa ggc ggc aag aag cag gct gag gca cag acc ggc atc acc     3016
Val Lys Glu Gly Gly Lys Lys Gln Ala Glu Ala Gln Thr Gly Ile Thr
        845                 850                 855

ggc tcc cag aag ctt cca cac ggc ctt gag acc aac atc tcc cgt gaa     3064
Gly Ser Gln Lys Leu Pro His Gly Leu Glu Thr Asn Ile Ser Arg Glu
    860                 865                 870

gag ctc ctg gaa ctg gga cag gct ttc gcc aac acc cca gaa ggc ttc     3112
Glu Leu Leu Glu Leu Gly Gln Ala Phe Ala Asn Thr Pro Glu Gly Phe
875                 880                 885                 890

aac tac cac cca cgt gtg gct cca gtt gct aag aag cgc gtc tcc tct     3160
Asn Tyr His Pro Arg Val Ala Pro Val Ala Lys Lys Arg Val Ser Ser
                895                 900                 905

gtc acc gaa ggt ggc atc gac tgg gca tgg ggc gag ctc ctc gcc ttc     3208
Val Thr Glu Gly Gly Ile Asp Trp Ala Trp Gly Glu Leu Leu Ala Phe
            910                 915                 920

ggt tcc ctg gct aac tcc ggc cgc ttg gtt cgc ctt gca ggt gaa gat     3256
Gly Ser Leu Ala Asn Ser Gly Arg Leu Val Arg Leu Ala Gly Glu Asp
        925                 930                 935

tcc cgc cgc ggt acc ttc acc cag cgc cac gca gtt gcc atc gac cca     3304
Ser Arg Arg Gly Thr Phe Thr Gln Arg His Ala Val Ala Ile Asp Pro
    940                 945                 950

gcg acc gct gaa gag ttc aac cca ctc cac gag ctt gca cag tcc aag     3352
Ala Thr Ala Glu Glu Phe Asn Pro Leu His Glu Leu Ala Gln Ser Lys
955                 960                 965                 970

ggc aac aac ggt aag ttc ctg gtc tac aac tcc gca ctg acc gag tac     3400
Gly Asn Asn Gly Lys Phe Leu Val Tyr Asn Ser Ala Leu Thr Glu Tyr
                975                 980                 985

gca ggc atg ggc ttc gag tac ggc tac tcc gta gga aac gaa gac tcc     3448
Ala Gly Met Gly Phe Glu Tyr Gly Tyr Ser Val Gly Asn Glu Asp Ser
            990                 995                 1000
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gtc gtt gca  tgg gaa gca cag ttc  ggc gac ttc gcc aac  ggc gct      3493
Val Val Ala  Trp Glu Ala Gln Phe  Gly Asp Phe Ala Asn  Gly Ala
        1005                 1010                 1015

cag acc atc  atc gat gag tac gtc  tcc tca ggc gaa gct  aag tgg      3538
Gln Thr Ile  Ile Asp Glu Tyr Val  Ser Ser Gly Glu Ala  Lys Trp
        1020                 1025                 1030

ggc cag acc  tcc aag ctg atc ctt  ctg ctg cct cac ggc  tac gaa      3583
Gly Gln Thr  Ser Lys Leu Ile Leu  Leu Leu Pro His Gly  Tyr Glu
        1035                 1040                 1045

ggc cag ggc  cca gac cac tct tcc  gca cgt atc gag cgc  ttc ctg      3628
Gly Gln Gly  Pro Asp His Ser Ser  Ala Arg Ile Glu Arg  Phe Leu
        1050                 1055                 1060

cag ctg tgc  gct gag ggt tcc atg  act gtt gct cag cca  tcc acc      3673
Gln Leu Cys  Ala Glu Gly Ser Met  Thr Val Ala Gln Pro  Ser Thr
        1065                 1070                 1075

cca gca aac  cac ttc cac ctg ctg  cgt cgt cac gct ctg  tcc gac      3718
Pro Ala Asn  His Phe His Leu Leu  Arg Arg His Ala Leu  Ser Asp
        1080                 1085                 1090

ctg aag cgt  cca ctg gtt atc ttc  acc ccg aag tcc atg  ctg cgt      3763
Leu Lys Arg  Pro Leu Val Ile Phe  Thr Pro Lys Ser Met  Leu Arg
        1095                 1100                 1105

aac aag gct  gct gcc tcc gca cca  gaa gac ttc act gag  gtc acc      3808
Asn Lys Ala  Ala Ala Ser Ala Pro  Glu Asp Phe Thr Glu  Val Thr
        1110                 1115                 1120

aag ttc caa  tcc gtg atc gac gat  cca aac gtt gca gat  gca gcc      3853
Lys Phe Gln  Ser Val Ile Asp Asp  Pro Asn Val Ala Asp  Ala Ala
        1125                 1130                 1135

aag gtg aag  aag gtc atg ctg gtc  tcc ggc aag ctg tac  tac gaa      3898
Lys Val Lys  Lys Val Met Leu Val  Ser Gly Lys Leu Tyr  Tyr Glu
        1140                 1145                 1150

ttg gca aag  cgc aag gag aag gac  gga cgc gac gac atc  gcg atc      3943
Leu Ala Lys  Arg Lys Glu Lys Asp  Gly Arg Asp Asp Ile  Ala Ile
        1155                 1160                 1165

gtt cgt atc  gaa atg ctc cac cca  att ccg ttc aac cgc  atc tcc      3988
Val Arg Ile  Glu Met Leu His Pro  Ile Pro Phe Asn Arg  Ile Ser
        1170                 1175                 1180

gag gct ctt  gcc ggc tac cct aac  gct gag gaa gtc ctc  ttc gtt      4033
Glu Ala Leu  Ala Gly Tyr Pro Asn  Ala Glu Glu Val Leu  Phe Val
        1185                 1190                 1195

cag gat gag  cca gca aac cag ggc  cca tgg ccg ttc tac  cag gag      4078
Gln Asp Glu  Pro Ala Asn Gln Gly  Pro Trp Pro Phe Tyr  Gln Glu
        1200                 1205                 1210

cac ctc cca  gag ctg atc ccg aac  atg cca aag atg cgc  cgc gtt      4123
His Leu Pro  Glu Leu Ile Pro Asn  Met Pro Lys Met Arg  Arg Val
        1215                 1220                 1225
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tcc cgc cgc  gct cag tcc tcc acc  gca act ggt gtt gct  aag gtg      4168
Ser Arg Arg  Ala Gln Ser Ser Thr  Ala Thr Gly Val Ala  Lys Val
        1230                1235                1240
cac cag ctg  gag gag aag cag ctt  atc gac gag gct ttc  gag gct      4213
His Gln Leu  Glu Glu Lys Gln Leu  Ile Asp Glu Ala Phe  Glu Ala
        1245                1250                1255
taagtcttta tagtcctgca ctagcctaga gggccttatg cagtgtgaat cacacagcat  4273
aaggcccttt ttgctgccgt ggttgcctaa ggtggaaggc atgaaacgaa tctgtgcggt  4333
cacgatctct tcagtacttt tgctaagtgg ctgctcctcc acttccacca cgcagctcga  4393
g                                                                  4394
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<210> 31
<211> 1257
<212> PRT
<213> Corynebacterium glutamicum

<400> 31

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1               5                  10                  15
Val Asp Lys Ile Lys Leu Asn Lys Pro Ser Arg Ser Lys Glu Lys Arg
            20                  25                  30
Arg Val Pro Ala Val Ser Ser Ala Ser Thr Phe Gly Gln Asn Ala Trp
        35                  40                  45
Leu Val Asp Glu Met Phe Gln Gln Phe Gln Lys Asp Pro Lys Ser Val
    50                  55                  60
Asp Lys Glu Trp Arg Glu Leu Phe Glu Ala Gln Gly Gly Pro Asn Ala
65                  70                  75                  80
Thr Pro Ala Thr Thr Glu Ala Gln Pro Ser Ala Pro Lys Glu Ser Ala
                85                  90                  95
Lys Pro Ala Pro Lys Ala Ala Pro Ala Ala Lys Ala Ala Pro Arg Val
            100                 105                 110
Glu Thr Lys Pro Ala Ala Lys Thr Ala Pro Lys Ala Lys Glu Ser Ser
        115                 120                 125
Val Pro Gln Gln Pro Lys Leu Pro Glu Pro Gly Gln Thr Pro Ile Arg
    130                 135                 140
Gly Ile Phe Lys Ser Ile Ala Lys Asn Met Asp Ile Ser Leu Glu Ile
145                 150                 155                 160
Pro Thr Ala Thr Ser Val Arg Asp Met Pro Ala Arg Leu Met Phe Glu
                165                 170                 175
Asn Arg Ala Met Val Asn Asp Gln Leu Lys Arg Thr Arg Gly Gly Lys
            180                 185                 190
Ile Ser Phe Thr His Ile Ile Gly Tyr Ala Met Val Lys Ala Val Met
        195                 200                 205
Ala His Pro Asp Met Asn Asn Ser Tyr Asp Val Ile Asp Gly Lys Pro
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    210                 215                 220
Thr Leu Ile Val Pro Glu His Ile Asn Leu Gly Leu Ala Ile Asp Leu
225                 230                 235                 240
Pro Gln Lys Asp Gly Ser Arg Ala Leu Val Val Ala Ala Ile Lys Glu
                245                 250                 255
Thr Glu Lys Met Asn Phe Ser Glu Phe Leu Ala Ala Tyr Glu Asp Ile
            260                 265                 270
Val Thr Arg Ser Arg Lys Gly Lys Leu Thr Met Asp Asp Tyr Gln Gly
        275                 280                 285
Val Thr Val Ser Leu Thr Asn Pro Gly Gly Ile Gly Thr Arg His Ser
    290                 295                 300
Val Pro Arg Leu Thr Lys Gly Gln Gly Thr Ile Ile Gly Val Gly Ser
305                 310                 315                 320
Met Asp Tyr Pro Ala Glu Phe Gln Gly Ala Ser Glu Asp Arg Leu Ala
                325                 330                 335
Glu Leu Gly Val Gly Lys Leu Val Thr Ile Thr Ser Thr Tyr Asp His
            340                 345                 350
Arg Val Ile Gln Gly Ala Val Ser Gly Glu Phe Leu Arg Thr Met Ser
        355                 360                 365
Arg Leu Leu Thr Asp Asp Ser Phe Trp Asp Glu Ile Phe Asp Ala Met
    370                 375                 380
Asn Val Pro Tyr Thr Pro Met Arg Trp Ala Gln Asp Val Pro Asn Thr
385                 390                 395                 400
Gly Val Asp Lys Asn Thr Arg Val Met Gln Leu Ile Glu Ala Tyr Arg
                405                 410                 415
Ser Arg Gly His Leu Ile Ala Asp Thr Asn Pro Leu Ser Trp Val Gln
            420                 425                 430
Pro Gly Met Pro Val Pro Asp His Arg Asp Leu Asp Ile Glu Thr His
        435                 440                 445
Ser Leu Thr Ile Trp Asp Leu Asp Arg Thr Phe Ser Val Gly Gly Phe
    450                 455                 460
Gly Gly Lys Glu Thr Met Thr Leu Arg Glu Val Leu Ser Arg Leu Arg
465                 470                 475                 480
Ala Ala Tyr Thr Leu Lys Val Gly Ser Glu Tyr Thr His Ile Leu Asp
                485                 490                 495
Arg Asp Glu Arg Thr Trp Leu Gln Asp Arg Leu Glu Ala Gly Met Pro
            500                 505                 510
Lys Pro Thr Gln Ala Glu Gln Lys Tyr Ile Leu Gln Lys Leu Asn Ala
        515                 520                 525
Ala Glu Ala Phe Glu Asn Phe Leu Gln Thr Lys Tyr Val Gly Gln Lys
    530                 535                 540
Arg Phe Ser Leu Glu Gly Ala Glu Ala Leu Ile Pro Leu Met Asp Ser
545                 550                 555                 560
Ala Ile Asp Thr Ala Ala Gly Gln Gly Leu Asp Glu Val Val Ile Gly
                565                 570                 575
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Met Pro His Arg Gly Arg Leu Asn Val Leu Phe Asn Ile Val Gly Lys
580 585 590
Pro Leu Ala Ser Ile Phe Asn Glu Phe Glu Gly Gln Met Glu Gln Gly
595 600 605
Gln Ile Gly Gly Ser Gly Asp Val Lys Tyr His Leu Gly Ser Glu Gly
610 615 620
Gln His Leu Gln Met Phe Gly Asp Gly Glu Ile Lys Val Ser Leu Thr
625 630 635 640
Ala Asn Pro Ser His Leu Glu Ala Val Asn Pro Val Met Glu Gly Ile
645 650 655
Val Arg Ala Lys Gln Asp Tyr Leu Asp Lys Gly Val Asp Gly Lys Thr
660 665 670
Val Val Pro Leu Leu Leu His Gly Asp Ala Ala Phe Ala Gly Leu Gly
675 680 685
Ile Val Pro Glu Thr Ile Asn Leu Ala Lys Leu Arg Gly Tyr Asp Val
690 695 700
Gly Gly Thr Ile His Ile Val Val Asn Asn Gln Ile Gly Phe Thr Thr
705 710 715 720
Thr Pro Asp Ser Ser Arg Ser Met His Tyr Ala Thr Asp Tyr Ala Lys
725 730 735
Ala Phe Gly Cys Pro Val Phe His Val Asn Gly Asp Asp Pro Glu Ala
740 745 750
Val Val Trp Val Gly Gln Leu Ala Thr Glu Tyr Arg Arg Arg Phe Gly
755 760 765
Lys Asp Val Phe Ile Asp Leu Val Cys Tyr Arg Leu Arg Gly His Asn
770 775 780
Glu Ala Asp Asp Pro Ser Met Thr Gln Pro Lys Met Tyr Glu Leu Ile
785 790 795 800
Thr Gly Arg Glu Thr Val Arg Ala Gln Tyr Thr Glu Asp Leu Leu Gly
805 810 815
Arg Gly Asp Leu Ser Asn Glu Asp Ala Glu Ala Val Val Arg Asp Phe
820 825 830
His Asp Gln Met Glu Ser Val Phe Asn Glu Val Lys Glu Gly Gly Lys
835 840 845
Lys Gln Ala Glu Ala Gln Thr Gly Ile Thr Gly Ser Gln Lys Leu Pro
850 855 860
His Gly Leu Glu Thr Asn Ile Ser Arg Glu Glu Leu Leu Glu Leu Gly
865 870 875 880
Gln Ala Phe Ala Asn Thr Pro Glu Gly Phe Asn Tyr His Pro Arg Val
885 890 895
Ala Pro Val Ala Lys Lys Arg Val Ser Ser Val Thr Glu Gly Gly Ile
900 905 910
Asp Trp Ala Trp Gly Glu Leu Leu Ala Phe Gly Ser Leu Ala Asn Ser
915 920 925
Gly Arg Leu Val Arg Leu Ala Gly Glu Asp Ser Arg Arg Gly Thr Phe

930 935 940

Thr Gln Arg His Ala Val Ala Ile Asp Pro Ala Thr Ala Glu Glu Phe
945 950 955 960
Asn Pro Leu His Glu Leu Ala Gln Ser Lys Gly Asn Asn Gly Lys Phe
965 970 975
Leu Val Tyr Asn Ser Ala Leu Thr Glu Tyr Ala Gly Met Gly Phe Glu
980 985 990
Tyr Gly Tyr Ser Val Gly Asn Glu Asp Ser Val Val Ala Trp Glu Ala
995 1000 1005
Gln Phe Gly Asp Phe Ala Asn Gly Ala Gln Thr Ile Ile Asp Glu
1010 1015 1020
Tyr Val Ser Ser Gly Glu Ala Lys Trp Gly Gln Thr Ser Lys Leu
1025 1030 1035
Ile Leu Leu Leu Pro His Gly Tyr Glu Gly Gln Gly Pro Asp His
1040 1045 1050
Ser Ser Ala Arg Ile Glu Arg Phe Leu Gln Leu Cys Ala Glu Gly
1055 1060 1065
Ser Met Thr Val Ala Gln Pro Ser Thr Pro Ala Asn His Phe His
1070 1075 1080
Leu Leu Arg Arg His Ala Leu Ser Asp Leu Lys Arg Pro Leu Val
1085 1090 1095
Ile Phe Thr Pro Lys Ser Met Leu Arg Asn Lys Ala Ala Ala Ser
1100 1105 1110
Ala Pro Glu Asp Phe Thr Glu Val Thr Lys Phe Gln Ser Val Ile
1115 1120 1125
Asp Asp Pro Asn Val Ala Asp Ala Ala Lys Val Lys Lys Val Met
1130 1135 1140
Leu Val Ser Gly Lys Leu Tyr Tyr Glu Leu Ala Lys Arg Lys Glu
1145 1150 1155
Lys Asp Gly Arg Asp Asp Ile Ala Ile Val Arg Ile Glu Met Leu
1160 1165 1170
His Pro Ile Pro Phe Asn Arg Ile Ser Glu Ala Leu Ala Gly Tyr
1175 1180 1185
Pro Asn Ala Glu Glu Val Leu Phe Val Gln Asp Glu Pro Ala Asn
1190 1195 1200
Gln Gly Pro Trp Pro Phe Tyr Gln Glu His Leu Pro Glu Leu Ile
1205 1210 1215
Pro Asn Met Pro Lys Met Arg Arg Val Ser Arg Arg Ala Gln Ser
1220 1225 1230
Ser Thr Ala Thr Gly Val Ala Lys Val His Gln Leu Glu Glu Lys
1235 1240 1245
Gln Leu Ile Asp Glu Ala Phe Glu Ala
1250 1255

<210> 32
<211> 3481
<212> DNA

<213> Corynebacterium glutamicum

<220>
<221> CDS
<222> (1437).. (3035)

<400> 32

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agtgtcaacg tcaccggtga cgcggtcatc aaagcgggag ccgatagcaa tcagcaggtc     120
gctgcgctgc agtgcaccaa cagcggacac agtgccatgc atgcctggca tacccatgtg     180
cagctcgtgg gactctggga aggttcccag cgccatcaat gtggtgacaa ctggaatgcc     240
ggtgtgctca gcgaacgcac gaagctcttc gtgggcatca gccttgataa cgccgccgcc     300
aacgtaaagg acaggcttct tagactcacc gatcagtttg acagcctgct caatctgtcg     360
agcatgcggt gttgaaactg ggcggtagcc tggcaggtcg atctttggtg gccagacgaa     420
atccaattca gcgttctgaa catccttggg gatatccact agaacaggac cagggcgacc     480
agtaatcgcg aggtggaatg cctcagccaa tgcctgtgga atgtcgttgg ggttggtgac     540
catgaagttg tgcttggtca ctggcatggt gatgccgcgg atatcggctt cctggaaagc     600
atcggtaccc agcaggctac ttccgacctg gccggtgatg gcaaccatgg gaacggagtc     660
caagtttgca tcagcgattg gggtaaccaa gttggttgcg cctgggccag aggttgcaat     720
gcagacgcca acgcgtccag taacctgcgc gtagccggtt gctgcgtggc ctgcgccctg     780
ctcgtggcgc actaggacgt ggcgcacctt tgtggaggaa tagagcgggt catacaccgg     840
tagcaccgca ccaccaggaa taccgaacac gatgtcggcg ttaagctcct cgagcgatcg     900
aacaattgcc tgtgcacctg tcatccgctc aggggcggcg gatcgaccac ggcttgcaac     960
cgtggcggga gtgggctgtt gagaagctgc cacattcacg actttctggc tcctttacta    1020
aataaggatt ttcacaggac ccgtccaagc caagccgatt tcaactcagc ctaaagacaa    1080
agccctcatt taaaattgtt ccgacgcgga tgcgtgtgca cgcagtgcga cagatgtctg    1140
ttgcaaagtt ggctacttgg gtcataacca acaagaaagc cctcgttcca acactgtggt    1200
gagtgttgtc gagggcgctt gacgagacga cttggaaggc cgttacggca ggcgccgcgc    1260
ggttactact acaagtcgaa taatggtcat ggtgtgtcat gctacacaca tcgagtttcc    1320
aattccacaa cgcacgaaaa ttcccacccc caaaactccc ccacttcggt taaggaatca    1380
ggattctcac aaagttcagg caggctcccg ctacttttca gcgctaatct tggctc atg    1439
                                                              Met
                                                               1

att tta ggc gta ccc att caa tat ttg ctc tat tca ttg tgg aat tgg      1487
Ile Leu Gly Val Pro Ile Gln Tyr Leu Leu Tyr Ser Leu Trp Asn Trp
            5                  10                  15

att gtc gat acc ggt ttt gat gta gca att atc ctg gtc ttg gcg ttt      1535
Ile Val Asp Thr Gly Phe Asp Val Ala Ile Ile Leu Val Leu Ala Phe
        20                  25                  30

ttg att cca cgt atc ggc cga ctg gcc atg cgt att atc aag cag cga      1583
Leu Ile Pro Arg Ile Gly Arg Leu Ala Met Arg Ile Ile Lys Gln Arg
    35                  40                  45
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gtg gag tct gca gcc gat gcg gac acc act aag aac cag ctc gcg ttc    1631
Val Glu Ser Ala Ala Asp Ala Asp Thr Thr Lys Asn Gln Leu Ala Phe
50                  55                  60                  65
gct ggc gtt ggc gtt tat atc gcg caa att gtg gcg ttt ttc atg ctt    1679
Ala Gly Val Gly Val Tyr Ile Ala Gln Ile Val Ala Phe Phe Met Leu
                70                  75                  80
gcc gtc tcc gcg atg cag gct ttt ggt ttc tct ctc gcg ggc gct gcg    1727
Ala Val Ser Ala Met Gln Ala Phe Gly Phe Ser Leu Ala Gly Ala Ala
            85                  90                  95
att ccg gca acc att gcg tca gct gcc att ggt ctt ggt gcg cag tcg    1775
Ile Pro Ala Thr Ile Ala Ser Ala Ala Ile Gly Leu Gly Ala Gln Ser
        100                 105                 110
att gtt gcg gac ttc ttg gcc gga ttt ttc atc ctg acg gaa aag caa    1823
Ile Val Ala Asp Phe Leu Ala Gly Phe Phe Ile Leu Thr Glu Lys Gln
    115                 120                 125
ttc ggc gtg ggt gac tgg gtg cgc ttt gag ggc aac ggc atc gtt gtt    1871
Phe Gly Val Gly Asp Trp Val Arg Phe Glu Gly Asn Gly Ile Val Val
130                 135                 140                 145
gaa ggc acc gtc att gag atc acc atg cgc gcg acc aaa att cgc acg    1919
Glu Gly Thr Val Ile Glu Ile Thr Met Arg Ala Thr Lys Ile Arg Thr
                150                 155                 160
att gca caa gag acc gtg atc atc ccg aac tcc acg gcg aaa gtg tgc    1967
Ile Ala Gln Glu Thr Val Ile Ile Pro Asn Ser Thr Ala Lys Val Cys
            165                 170                 175
atc aac aat tct aat aac tgg tcg cgt gcg gtt gtc gtt att ccg atc    2015
Ile Asn Asn Ser Asn Asn Trp Ser Arg Ala Val Val Val Ile Pro Ile
        180                 185                 190
ccc atg ttg ggt tct gaa aac atc aca gat gtc atc gcg cgc tct gaa    2063
Pro Met Leu Gly Ser Glu Asn Ile Thr Asp Val Ile Ala Arg Ser Glu
    195                 200                 205
gct gcg act cgt cgc gca ctt ggc cag gag aaa atc gca ccg gaa atc    2111
Ala Ala Thr Arg Arg Ala Leu Gly Gln Glu Lys Ile Ala Pro Glu Ile
210                 215                 220                 225
ctc ggt gaa ctc gat gtg cac cca gcc acg gaa gtc aca ccg cca acg    2159
Leu Gly Glu Leu Asp Val His Pro Ala Thr Glu Val Thr Pro Pro Thr
                230                 235                 240
gtg gtc ggc atg ccg tgg atg gtc acc atg cgt ttc ctc gtg caa gtc    2207
Val Val Gly Met Pro Trp Met Val Thr Met Arg Phe Leu Val Gln Val
            245                 250                 255
acc gcc ggc aat caa tgg ctg gtc gaa cgc gcc atc cgc aca gaa atc    2255
Thr Ala Gly Asn Gln Trp Leu Val Glu Arg Ala Ile Arg Thr Glu Ile
        260                 265                 270
atc aac gaa ttc tgg gaa gaa tac ggc agc gca acc act aca tcg gga    2303
Ile Asn Glu Phe Trp Glu Glu Tyr Gly Ser Ala Thr Thr Thr Ser Gly
    275                 280                 285
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acc ctc att gat tcc tta cac gtt gag cat gaa gag cca aag acc tcg      2351
Thr Leu Ile Asp Ser Leu His Val Glu His Glu Glu Pro Lys Thr Ser
290                 295                 300                 305
ctt atc gac gcc tcc ccc cag gct ctt aag gaa ccg aag ccg gag gct      2399
Leu Ile Asp Ala Ser Pro Gln Ala Leu Lys Glu Pro Lys Pro Glu Ala
                310                 315                 320
gcg gcg acg gtt gca tcg cta gct gca tcg tct aac gac gat gca gac      2447
Ala Ala Thr Val Ala Ser Leu Ala Ala Ser Ser Asn Asp Asp Ala Asp
            325                 330                 335
aat gca gac gcc tcg gcg atc aat gca ggc aat cca gag aag gaa ctt      2495
Asn Ala Asp Ala Ser Ala Ile Asn Ala Gly Asn Pro Glu Lys Glu Leu
        340                 345                 350
gat tcc gat gtg ctg gaa caa gaa ctc tcc agc gaa gaa ccg gaa gaa      2543
Asp Ser Asp Val Leu Glu Gln Glu Leu Ser Ser Glu Glu Pro Glu Glu
    355                 360                 365
aca gca aaa cca gat cac tct ctc cga ggc ttc ttc cgc act gat tac      2591
Thr Ala Lys Pro Asp His Ser Leu Arg Gly Phe Phe Arg Thr Asp Tyr
370                 375                 380                 385
tac cca aat cgg tgg cag aag atc ctg tcg ttt ggc gga cgt gtc cgc      2639
Tyr Pro Asn Arg Trp Gln Lys Ile Leu Ser Phe Gly Gly Arg Val Arg
                390                 395                 400
atg agc act tcc ctg ttg ttg ggt gcg ctg ctc ttg ctg tca cta ttt      2687
Met Ser Thr Ser Leu Leu Leu Gly Ala Leu Leu Leu Leu Ser Leu Phe
            405                 410                 415
aag gtc atg act gtg gaa cca agt gag aat tgg caa aac tcc agt gga      2735
Lys Val Met Thr Val Glu Pro Ser Glu Asn Trp Gln Asn Ser Ser Gly
        420                 425                 430
tgg ctg tca cca agc act gcc acc tca act gcg gtg acc acc tcc gaa      2783
Trp Leu Ser Pro Ser Thr Ala Thr Ser Thr Ala Val Thr Thr Ser Glu
    435                 440                 445
act tcc gcg cca gca agc acg cct tcg atg aca gtg ccc act acg gtg      2831
Thr Ser Ala Pro Ala Ser Thr Pro Ser Met Thr Val Pro Thr Thr Val
450                 455                 460                 465
gag gag acc cca acg atg gaa tct agc gtc gaa acg cag cag gaa acc      2879
Glu Glu Thr Pro Thr Met Glu Ser Ser Val Glu Thr Gln Gln Glu Thr
                470                 475                 480
tca acc cct gca acc gca acg ccc cag cga gcc gac acc atc gaa ccg      2927
Ser Thr Pro Ala Thr Ala Thr Pro Gln Arg Ala Asp Thr Ile Glu Pro
            485                 490                 495
acc gag gaa gcc acg tcg cag gag gaa acg act gca tcg cag acg cag      2975
Thr Glu Glu Ala Thr Ser Gln Glu Glu Thr Thr Ala Ser Gln Thr Gln
        500                 505                 510
tct cca gca gtg gaa gca cca acc gcg gtc caa gaa aca gtt gcg ccg      3023
Ser Pro Ala Val Glu Ala Pro Thr Ala Val Gln Glu Thr Val Ala Pro
    515                 520                 525
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acg tcc acc cct taggacgctg attacagacg tgtcccattt ctttactact 3075
Thr Ser Thr Pro
530
attggaaatt atgagttcag acgcagaaaa ggcatccgtg gagctttccg aaaaatttca 3135
cccagaacgc acccatattt tgggcgccgt tgtttttggc ctgatctcat tattagtcat 3195
cggcgcagcc cctcagtacc tgttttggct gctcgcgctc cctgtcatct tcggttactg 3255
ggttctaaaa tcatccacga tcgttgatga acagggcatc accgcaaact acgccttcaa 3315
gggcaaaaag gttgtggcct gggaagacct cgcaggaatc ggattcaagg gtgcccgcac 3375
tttcgctcgc accacctccg atgcagaagt caccctcccc ggcgtcacct tcaactccct 3435
tccccgcctt gaagctgctt cccacggccg catccccgat gcgatc 3481

<210> 33
<211> 533
<212> PRT
<213> Corynebacterium glutamicum

<400> 33

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Trp Ile Val Asp Thr Gly Phe Asp Val Ala Ile Ile Leu Val Leu Ala
20 25 30
Phe Leu Ile Pro Arg Ile Gly Arg Leu Ala Met Arg Ile Ile Lys Gln
35 40 45
Arg Val Glu Ser Ala Ala Asp Ala Asp Thr Thr Lys Asn Gln Leu Ala
50 55 60
Phe Ala Gly Val Gly Val Tyr Ile Ala Gln Ile Val Ala Phe Phe Met
65 70 75 80
Leu Ala Val Ser Ala Met Gln Ala Phe Gly Phe Ser Leu Ala Gly Ala
85 90 95
Ala Ile Pro Ala Thr Ile Ala Ser Ala Ala Ile Gly Leu Gly Ala Gln
100 105 110
Ser Ile Val Ala Asp Phe Leu Ala Gly Phe Phe Ile Leu Thr Glu Lys
115 120 125
Gln Phe Gly Val Gly Asp Trp Val Arg Phe Glu Gly Asn Gly Ile Val
130 135 140
Val Glu Gly Thr Val Ile Glu Ile Thr Met Arg Ala Thr Lys Ile Arg
145 150 155 160
Thr Ile Ala Gln Glu Thr Val Ile Ile Pro Asn Ser Thr Ala Lys Val
165 170 175
Cys Ile Asn Asn Ser Asn Asn Trp Ser Arg Ala Val Val Val Ile Pro
180 185 190
Ile Pro Met Leu Gly Ser Glu Asn Ile Thr Asp Val Ile Ala Arg Ser
195 200 205
Glu Ala Ala Thr Arg Arg Ala Leu Gly Gln Glu Lys Ile Ala Pro Glu

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    210                 215                 220
Ile Leu Gly Glu Leu Asp Val His Pro Ala Thr Glu Val Thr Pro Pro
225                 230                 235                 240
Thr Val Val Gly Met Pro Trp Met Val Thr Met Arg Phe Leu Val Gln
                245                 250                 255
Val Thr Ala Gly Asn Gln Trp Leu Val Glu Arg Ala Ile Arg Thr Glu
            260                 265                 270
Ile Ile Asn Glu Phe Trp Glu Glu Tyr Gly Ser Ala Thr Thr Thr Ser
        275                 280                 285
Gly Thr Leu Ile Asp Ser Leu His Val Glu His Glu Glu Pro Lys Thr
    290                 295                 300
Ser Leu Ile Asp Ala Ser Pro Gln Ala Leu Lys Glu Pro Lys Pro Glu
305                 310                 315                 320
Ala Ala Ala Thr Val Ala Ser Leu Ala Ala Ser Ser Asn Asp Asp Ala
                325                 330                 335
Asp Asn Ala Asp Ala Ser Ala Ile Asn Ala Gly Asn Pro Glu Lys Glu
            340                 345                 350
Leu Asp Ser Asp Val Leu Glu Gln Glu Leu Ser Ser Glu Glu Pro Glu
        355                 360                 365
Glu Thr Ala Lys Pro Asp His Ser Leu Arg Gly Phe Phe Arg Thr Asp
    370                 375                 380
Tyr Tyr Pro Asn Arg Trp Gln Lys Ile Leu Ser Phe Gly Gly Arg Val
385                 390                 395                 400
Arg Met Ser Thr Ser Leu Leu Leu Gly Ala Leu Leu Leu Leu Ser Leu
                405                 410                 415
Phe Lys Val Met Thr Val Glu Pro Ser Glu Asn Trp Gln Asn Ser Ser
            420                 425                 430
Gly Trp Leu Ser Pro Ser Thr Ala Thr Ser Thr Ala Val Thr Thr Ser
        435                 440                 445
Glu Thr Ser Ala Pro Ala Ser Thr Pro Ser Met Thr Val Pro Thr Thr
    450                 455                 460
Val Glu Glu Thr Pro Thr Met Glu Ser Ser Val Glu Thr Gln Gln Glu
465                 470                 475                 480
Thr Ser Thr Pro Ala Thr Ala Thr Pro Gln Arg Ala Asp Thr Ile Glu
                485                 490                 495
Pro Thr Glu Glu Ala Thr Ser Gln Glu Glu Thr Thr Ala Ser Gln Thr
            500                 505                 510
Gln Ser Pro Ala Val Glu Ala Pro Thr Ala Val Gln Glu Thr Val Ala
        515                 520                 525
Pro Thr Ser Thr Pro
    530
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<210> 34
<211> 2644
<212> DNA
<213> Corynebacterium glutamicum

<220>
<221> CDS
<222> (1214)..(2641)

<400> 34

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ttccttcagt tccaacggag tgaacttcgc ctgagcaggt ccacgacgac caaaaacgtg     180
cacttcctta gcctgattct tagccaagct ctcatagaca ttgtcaggga tttcagtaac     240
tagcagctca tcgccagtct tcgccaaaat acgagcaacg tccaacgcca cgttaccgac     300
accaacaacc gctaccttct cagcagaaag atcccagttg cgttcaaagt tcggattgcc     360
atcatagaaa ccaacgaact cgccagcgcc ccacgaacct tccagatcag aacctggaac     420
ccgaagatcc tggtcgccag tagcgccagt ggagaacacg atcgcgtcat aaaactcacg     480
caactcctca acagtgatgt ccttgccgac ctcaatgttg cccaagaaac gcagctgctc     540
cttgtccatc acattgtgca gggacttcac gatgcccttg atgcgagggt gatcaggcgc     600
aacaccataa cggatcaaac cgaaaggcgc tggcatgcgt tcaaaaagat caatctgcac     660
gtccgtgtcg gatttcatca acaaatcaga cgcgtagatt cctgctggac ctgcaccgac     720
aacggcaaca cgcaaagggc gagacatata aagttcgatt ccttaaaggg gttctaaaaa     780
atgtggagta tgtgagcggg gttccactag tagattcgac tcctatcggg gtgcgactgc     840
taatggtgcc ctgctatcaa ccctccatga tacgtggtaa gtgcagacta ataaaggcca     900
gtcggggagt attgggggct ttgctggggg cagatttgtc acgctgcgcg ctttcataga     960
ccccattaat gtggggtgaa gagctgtaaa gtaccgctaa aaactttgca aagggtgctt    1020
cgcaacttgt aaccgctccg tattgtttтc tacggcaata agcatttgtg ctgctcaaag    1080
cgtggaattg agatcggttt gaaaattaca aaataaaact ttgcaaaccg ggctgtacgc    1140
aaggcggacg aacgctaaac tatgtaagaa atcacaactt cccctcagta gtgccaggag    1200
gcacaagcct gaa gtg tca tca atg aga agg ttc agg ctg aaa cta gaa        1249
                Val Ser Ser Met Arg Arg Phe Arg Leu Lys Leu Glu
                1               5                   10
agg cga tgt atg tct gac aca ccg acc tca gct ctg atc acc acg gtc      1297
Arg Arg Cys Met Ser Asp Thr Pro Thr Ser Ala Leu Ile Thr Thr Val
        15                  20                  25
aac cgc agc ttc gat gga ttc gat ttg gaa gaa gta gca gca gac ctt      1345
Asn Arg Ser Phe Asp Gly Phe Asp Leu Glu Glu Val Ala Ala Asp Leu
    30                  35                  40
gga gtt cgg ctc acc gac ctg ccc gac gaa gaa cta gaa gta tcc aaa      1393
Gly Val Arg Leu Thr Asp Leu Pro Asp Glu Glu Leu Glu Val Ser Lys
45                  50                  55                  60
gtt ctc gcg gcg gac ctc ctc gct gag ggg cca gct ctc atc atc ggt      1441
Val Leu Ala Ala Asp Leu Leu Ala Glu Gly Pro Ala Leu Ile Ile Gly
                65                  70                  75
gta gga aac acg ttt ttc gac gcc cag gtc gcc gct gcc ctc ggc gtc      1489
```

```
Val Gly Asn Thr Phe Phe Asp Ala Gln Val Ala Ala Ala Leu Gly Val
                80                  85                  90
cca gtg cta ctg ctg gta gac aag caa ggc aag cac gtt gct ctt gct  1537
Pro Val Leu Leu Leu Val Asp Lys Gln Gly Lys His Val Ala Leu Ala
        95                 100                 105
cgc acc cag gta aac aat gcc ggc gca gtt gtt gca gca gca ttt acc  1585
Arg Thr Gln Val Asn Asn Ala Gly Ala Val Val Ala Ala Ala Phe Thr
    110                 115                 120
gct gaa caa gag cca atg ccg gat aag ctg cgc aag gct gtg cgc aac  1633
Ala Glu Gln Glu Pro Met Pro Asp Lys Leu Arg Lys Ala Val Arg Asn
125                 130                 135                 140
cac agc aac ctc gaa cca gtc atg agc gcc gaa ctc ttt gaa aac tgg  1681
His Ser Asn Leu Glu Pro Val Met Ser Ala Glu Leu Phe Glu Asn Trp
                145                 150                 155
ctg ctc aag cgc gca cgc gca gag cac tcc cac att gtg ctg cca gaa  1729
Leu Leu Lys Arg Ala Arg Ala Glu His Ser His Ile Val Leu Pro Glu
            160                 165                 170
ggt gac gac gac cgc atc ttg atg gct gcc cac cag ctg ctt gat caa  1777
Gly Asp Asp Asp Arg Ile Leu Met Ala Ala His Gln Leu Leu Asp Gln
        175                 180                 185
gac atc tgt gac atc acg atc ctg ggc gat cca gta cag atc aag gag  1825
Asp Ile Cys Asp Ile Thr Ile Leu Gly Asp Pro Val Gln Ile Lys Glu
    190                 195                 200
cgc gct acc gaa ctt ggc ctg cac ctt aac act gca tac ctg gtc aat  1873
Arg Ala Thr Glu Leu Gly Leu His Leu Asn Thr Ala Tyr Leu Val Asn
205                 210                 215                 220
ccg ctg aca gat cct cgc ctg gag gaa ttc gcc gaa caa ttc gcg gag  1921
Pro Leu Thr Asp Pro Arg Leu Glu Glu Phe Ala Glu Gln Phe Ala Glu
                225                 230                 235
ctg cgc aag tca aag agc gtc act atc gat gaa gcc cgc gaa atc atg  1969
Leu Arg Lys Ser Lys Ser Val Thr Ile Asp Glu Ala Arg Glu Ile Met
            240                 245                 250
aag gat att tgc tac ttc ggc acc atg atg gtc cac aac ggc gac gcc  2017
Lys Asp Ile Cys Tyr Phe Gly Thr Met Met Val His Asn Gly Asp Ala
        255                 260                 265
gac gga atg gta tcc ggt gca gca aac acc acc gca cac acc att aag  2065
Asp Gly Met Val Ser Gly Ala Ala Asn Thr Thr Ala His Thr Ile Lys
    270                 275                 280
cca agc ttc cag atc atc aaa act gtt cca gaa gca tcc gtc gtt tct  2113
Pro Ser Phe Gln Ile Ile Lys Thr Val Pro Glu Ala Ser Val Val Ser
285                 290                 295                 300
tcc atc ttc ctc atg gtg ctg cgc ggg cga ctg tgg gca ttc ggc gac  2161
Ser Ile Phe Leu Met Val Leu Arg Gly Arg Leu Trp Ala Phe Gly Asp
                305                 310                 315
tgt gct gtt aac ccg aac cca act gct gaa cag ctt ggt gaa atc gcc  2209
```

```
Cys Ala Val Asn Pro Asn Pro Thr Ala Glu Gln Leu Gly Glu Ile Ala
            320                 325                 330
gtt gtg tca gca aaa act gca gca caa ttt ggc att gat cct cgc gta    2257
Val Val Ser Ala Lys Thr Ala Ala Gln Phe Gly Ile Asp Pro Arg Val
        335                 340                 345
gcc atc ttg tcc tac tcc act ggc aac tcc ggc gga ggc tca gat gtg    2305
Ala Ile Leu Ser Tyr Ser Thr Gly Asn Ser Gly Gly Gly Ser Asp Val
    350                 355                 360
gat cgc gcc atc gac gct ctt gca gaa gca cgc cga ctt aac cca gaa    2353
Asp Arg Ala Ile Asp Ala Leu Ala Glu Ala Arg Arg Leu Asn Pro Glu
365                 370                 375                 380
cta tgc gtc gat gga cca ctt cag ttc gac gcc gcc gtc gac ccg ggt    2401
Leu Cys Val Asp Gly Pro Leu Gln Phe Asp Ala Ala Val Asp Pro Gly
                385                 390                 395
gtg gcg cgc aag aag atg cca gac tct gac gtc gct ggc cag gca aat    2449
Val Ala Arg Lys Lys Met Pro Asp Ser Asp Val Ala Gly Gln Ala Asn
            400                 405                 410
gtg ttt atc ttc cct gac ctg gaa gcc gga aac atc ggc tac aaa act    2497
Val Phe Ile Phe Pro Asp Leu Glu Ala Gly Asn Ile Gly Tyr Lys Thr
        415                 420                 425
gca caa cgc acc ggt cac gcc ctg gca gtt ggt ccg att ctg cag ggc    2545
Ala Gln Arg Thr Gly His Ala Leu Ala Val Gly Pro Ile Leu Gln Gly
    430                 435                 440
cta aac aaa cca gtc aac gac ctt tcc cgt ggc gca aca gtc cct gac    2593
Leu Asn Lys Pro Val Asn Asp Leu Ser Arg Gly Ala Thr Val Pro Asp
445                 450                 455                 460
atc gtc aac aca gta gcc atc aca gca att cag gca gga gga cgc agc    2641
Ile Val Asn Thr Val Ala Ile Thr Ala Ile Gln Ala Gly Gly Arg Ser
                465                 470                 475
taa                                                                2644
```

<210> 35
<211> 476
<212> PRT
<213> Corynebacterium glutamicum

<400> 35

```
Val Ser Ser Met Arg Arg Phe Arg Leu Lys Leu Glu Arg Arg Cys Met
1               5                  10                  15
Ser Asp Thr Pro Thr Ser Ala Leu Ile Thr Thr Val Asn Arg Ser Phe
            20                  25                  30
Asp Gly Phe Asp Leu Glu Glu Val Ala Ala Asp Leu Gly Val Arg Leu
        35                  40                  45
Thr Asp Leu Pro Asp Glu Glu Leu Glu Val Ser Lys Val Leu Ala Ala
```

```
    50                  55                  60
Asp Leu Leu Ala Glu Gly Pro Ala Leu Ile Ile Gly Val Gly Asn Thr
65                  70                  75                  80
Phe Phe Asp Ala Gln Val Ala Ala Ala Leu Gly Val Pro Val Leu Leu
                85                  90                  95
Leu Val Asp Lys Gln Gly Lys His Val Ala Leu Ala Arg Thr Gln Val
            100                 105                 110
Asn Asn Ala Gly Ala Val Val Ala Ala Ala Phe Thr Ala Glu Gln Glu
        115                 120                 125
Pro Met Pro Asp Lys Leu Arg Lys Ala Val Arg Asn His Ser Asn Leu
    130                 135                 140
Glu Pro Val Met Ser Ala Glu Leu Phe Glu Asn Trp Leu Leu Lys Arg
145                 150                 155                 160
Ala Arg Ala Glu His Ser His Ile Val Leu Pro Glu Gly Asp Asp Asp
                165                 170                 175
Arg Ile Leu Met Ala Ala His Gln Leu Leu Asp Gln Asp Ile Cys Asp
            180                 185                 190
Ile Thr Ile Leu Gly Asp Pro Val Gln Ile Lys Glu Arg Ala Thr Glu
        195                 200                 205
Leu Gly Leu His Leu Asn Thr Ala Tyr Leu Val Asn Pro Leu Thr Asp
    210                 215                 220
Pro Arg Leu Glu Glu Phe Ala Glu Gln Phe Ala Glu Leu Arg Lys Ser
225                 230                 235                 240
Lys Ser Val Thr Ile Asp Glu Ala Arg Glu Ile Met Lys Asp Ile Cys
                245                 250                 255
Tyr Phe Gly Thr Met Met Val His Asn Gly Asp Ala Asp Gly Met Val
            260                 265                 270
Ser Gly Ala Ala Asn Thr Thr Ala His Thr Ile Lys Pro Ser Phe Gln
        275                 280                 285
Ile Ile Lys Thr Val Pro Glu Ala Ser Val Val Ser Ser Ile Phe Leu
    290                 295                 300
Met Val Leu Arg Gly Arg Leu Trp Ala Phe Gly Asp Cys Ala Val Asn
305                 310                 315                 320
Pro Asn Pro Thr Ala Glu Gln Leu Gly Glu Ile Ala Val Val Ser Ala
                325                 330                 335
Lys Thr Ala Ala Gln Phe Gly Ile Asp Pro Arg Val Ala Ile Leu Ser
            340                 345                 350
Tyr Ser Thr Gly Asn Ser Gly Gly Gly Ser Asp Val Asp Arg Ala Ile
        355                 360                 365
Asp Ala Leu Ala Glu Ala Arg Arg Leu Asn Pro Glu Leu Cys Val Asp
    370                 375                 380
Gly Pro Leu Gln Phe Asp Ala Ala Val Asp Pro Gly Val Ala Arg Lys
385                 390                 395                 400
Lys Met Pro Asp Ser Asp Val Ala Gly Gln Ala Asn Val Phe Ile Phe
                405                 410                 415
```

```
Pro Asp Leu Glu Ala Gly Asn Ile Gly Tyr Lys Thr Ala Gln Arg Thr
            420                 425                 430
Gly His Ala Leu Ala Val Gly Pro Ile Leu Gln Gly Leu Asn Lys Pro
        435                 440                 445
Val Asn Asp Leu Ser Arg Gly Ala Thr Val Pro Asp Ile Val Asn Thr
    450                 455                 460
Val Ala Ile Thr Ala Ile Gln Ala Gly Gly Arg Ser
465                 470                 475
```

<210> 36
<211> 1425
<212> DNA
<213> Corynebacterium glutamicum

<220>
<221> CDS
<222> (1).. (1422)

<400> 36

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atg gga aag aca tat gtg ggg tcc agg ctg cgc caa ctg cgc cgc gaa      48
Met Gly Lys Thr Tyr Val Gly Ser Arg Leu Arg Gln Leu Arg Arg Glu
1               5                   10                  15
aga gac ctg agc cag gcg tcg ttg gca gca acc ctt ggc tta tct gcc      96
Arg Asp Leu Ser Gln Ala Ser Leu Ala Ala Thr Leu Gly Leu Ser Ala
            20                  25                  30
agt tat gta aat cag att gag cac gac gta cgc ccg ctc acc gta ccg     144
Ser Tyr Val Asn Gln Ile Glu His Asp Val Arg Pro Leu Thr Val Pro
        35                  40                  45
gtg tta ttg cgc atc acc gag gcg ttc ggc gta gac gca acg ttt ttc     192
Val Leu Leu Arg Ile Thr Glu Ala Phe Gly Val Asp Ala Thr Phe Phe
    50                  55                  60
tcc cgc gac gat gat tcc cgc ctt tta gct gaa gtt cag gat gtc atg     240
Ser Arg Asp Asp Asp Ser Arg Leu Leu Ala Glu Val Gln Asp Val Met
65                  70                  75                  80
ttg gac cgg gag atc aac ccc gcg aac gtg gag ctc caa gag cta tcg     288
Leu Asp Arg Glu Ile Asn Pro Ala Asn Val Glu Leu Gln Glu Leu Ser
                85                  90                  95
gag atg gtg tac aac cac ccg cag ctg gcg cgc gcc atg gtg gaa atg     336
Glu Met Val Tyr Asn His Pro Gln Leu Ala Arg Ala Met Val Glu Met
            100                 105                 110
cac cag cgc tac cga aac gta cgc gac aag ttc tcc atc gca gtg gac     384
His Gln Arg Tyr Arg Asn Val Arg Asp Lys Phe Ser Ile Ala Val Asp
        115                 120                 125
```

```
aat cgc acc aac acg cct gag gaa cgt cgc ccc atc gcg gag gcc gtg      432
Asn Arg Thr Asn Thr Pro Glu Glu Arg Arg Pro Ile Ala Glu Ala Val
    130                 135                 140
agc atg ccg cac gaa gag gtg cgc gat ttc att tac gcc cgc caa aac      480
Ser Met Pro His Glu Glu Val Arg Asp Phe Ile Tyr Ala Arg Gln Asn
145                 150                 155                 160
tac ttc gat gcg ctc gac cgc cgc gcc gaa gcc atc gcc gcg caa ctg      528
Tyr Phe Asp Ala Leu Asp Arg Arg Ala Glu Ala Ile Ala Ala Gln Leu
                165                 170                 175
ggc tgg cag ccg tac gat tcc cgc gcc atg gaa gat tcg atc gcc agg      576
Gly Trp Gln Pro Tyr Asp Ser Arg Ala Met Glu Asp Ser Ile Ala Arg
            180                 185                 190
cgc ctg caa atg gat cac gat gtc acc atc acc tcc tcc aaa gag gaa      624
Arg Leu Gln Met Asp His Asp Val Thr Ile Thr Ser Ser Lys Glu Glu
        195                 200                 205
tcc ggc acg ctg cac cac ttt gac ccc gag acg cgc cta ctg aca atc      672
Ser Gly Thr Leu His His Phe Asp Pro Glu Thr Arg Leu Leu Thr Ile
    210                 215                 220
cac gca cgc ctc aac ccc gga caa cgc gcc ttc cgc atg gcc acc gaa      720
His Ala Arg Leu Asn Pro Gly Gln Arg Ala Phe Arg Met Ala Thr Glu
225                 230                 235                 240
ctc ggc tac cta gaa gcc aac gac ctc atc gaa ggc atc gtc gac gac      768
Leu Gly Tyr Leu Glu Ala Asn Asp Leu Ile Glu Gly Ile Val Asp Asp
                245                 250                 255
ggc atc tgg tcc acc ccc gaa gcc cgc acc cta gcc atc cgc ggc gtc      816
Gly Ile Trp Ser Thr Pro Glu Ala Arg Thr Leu Ala Ile Arg Gly Val
            260                 265                 270
gcc tcc tac ttc gcc gcc gcc gta atg ctg ccc tac aaa atc ttc cac      864
Ala Ser Tyr Phe Ala Ala Ala Val Met Leu Pro Tyr Lys Ile Phe His
        275                 280                 285
tcc gag gcc gaa aaa tcc ggc tac gac atc gaa tac cta ggt cag ctt      912
Ser Glu Ala Glu Lys Ser Gly Tyr Asp Ile Glu Tyr Leu Gly Gln Leu
    290                 295                 300
ttc ggc gtg ggc tac gag acg acc gcc cac cgc ctg tcc acc ctg cag      960
Phe Gly Val Gly Tyr Glu Thr Thr Ala His Arg Leu Ser Thr Leu Gln
305                 310                 315                 320
cgt ccc aac ctg cgc ggc atc ccg ttt acc ttc gtg cgc gtc gac cgc     1008
Arg Pro Asn Leu Arg Gly Ile Pro Phe Thr Phe Val Arg Val Asp Arg
                325                 330                 335
gcc ggc aac atg tcc aaa cgc caa tcc gcc acc ggc ttc cac ttc acc     1056
Ala Gly Asn Met Ser Lys Arg Gln Ser Ala Thr Gly Phe His Phe Thr
            340                 345                 350
cac tac ggc ggc acc tgc ccc ctg tgg aac gtg ttt gaa acc ttc acc     1104
His Tyr Gly Gly Thr Cys Pro Leu Trp Asn Val Phe Glu Thr Phe Thr
        355                 360                 365
```

```
aac ccc ggc caa gtg ctc cgc caa ttc gcg caa atg ccc gac gga cgc      1152
Asn Pro Gly Gln Val Leu Arg Gln Phe Ala Gln Met Pro Asp Gly Arg
    370                 375                 380
aac tac ctg tgg atc tca cgc acc gtg cga cac cac gaa gcc cgg ttc      1200
Asn Tyr Leu Trp Ile Ser Arg Thr Val Arg His His Glu Ala Arg Phe
385                 390                 395                 400
ggc gaa gtg gac aaa atg ttc gcc atc ggg ctc ggc tgc gaa gcg cgc      1248
Gly Glu Val Asp Lys Met Phe Ala Ile Gly Leu Gly Cys Glu Ala Arg
                405                 410                 415
cac gcc gac cgc acc gtg tac tcc cgc ggc ttc aac ctc cag gac cta      1296
His Ala Asp Arg Thr Val Tyr Ser Arg Gly Phe Asn Leu Gln Asp Leu
            420                 425                 430
tcc acc gcc acc ccc atc ggg tcc ggc tgc cga gtg tgc acc cgc gag      1344
Ser Thr Ala Thr Pro Ile Gly Ser Gly Cys Arg Val Cys Thr Arg Glu
        435                 440                 445
aac tgc gcg cag cgc gca ttc ccg tcc gtg cac ggc cgc atc aac atc      1392
Asn Cys Ala Gln Arg Ala Phe Pro Ser Val His Gly Arg Ile Asn Ile
    450                 455                 460
gac gcg cac gag tcc act atc gcg ccg tac taa                         1425
Asp Ala His Glu Ser Thr Ile Ala Pro Tyr
465                 470
```

<210> 37
<211> 474
<212> PRT
<213> Corynebacterium glutamicum

<400> 37

```
Met Gly Lys Thr Tyr Val Gly Ser Arg Leu Arg Gln Leu Arg Arg Glu
1               5                   10                  15
Arg Asp Leu Ser Gln Ala Ser Leu Ala Ala Thr Leu Gly Leu Ser Ala
            20                  25                  30
Ser Tyr Val Asn Gln Ile Glu His Asp Val Arg Pro Leu Thr Val Pro
        35                  40                  45
Val Leu Leu Arg Ile Thr Glu Ala Phe Gly Val Asp Ala Thr Phe Phe
    50                  55                  60
Ser Arg Asp Asp Asp Ser Arg Leu Leu Ala Glu Val Gln Asp Val Met
65                  70                  75                  80
Leu Asp Arg Glu Ile Asn Pro Ala Asn Val Glu Leu Gln Glu Leu Ser
                85                  90                  95
Glu Met Val Tyr Asn His Pro Gln Leu Ala Arg Ala Met Val Glu Met
            100                 105                 110
His Gln Arg Tyr Arg Asn Val Arg Asp Lys Phe Ser Ile Ala Val Asp
        115                 120                 125
```

```
Asn Arg Thr Asn Thr Pro Glu Glu Arg Arg Pro Ile Ala Glu Ala Val
    130                 135                 140
Ser Met Pro His Glu Glu Val Arg Asp Phe Ile Tyr Ala Arg Gln Asn
145                 150                 155                 160
Tyr Phe Asp Ala Leu Asp Arg Arg Ala Glu Ala Ile Ala Ala Gln Leu
                165                 170                 175
Gly Trp Gln Pro Tyr Asp Ser Arg Ala Met Glu Asp Ser Ile Ala Arg
            180                 185                 190
Arg Leu Gln Met Asp His Asp Val Thr Ile Thr Ser Ser Lys Glu Glu
        195                 200                 205
Ser Gly Thr Leu His His Phe Asp Pro Glu Thr Arg Leu Leu Thr Ile
    210                 215                 220
His Ala Arg Leu Asn Pro Gly Gln Arg Ala Phe Arg Met Ala Thr Glu
225                 230                 235                 240
Leu Gly Tyr Leu Glu Ala Asn Asp Leu Ile Glu Gly Ile Val Asp Asp
                245                 250                 255
Gly Ile Trp Ser Thr Pro Glu Ala Arg Thr Leu Ala Ile Arg Gly Val
            260                 265                 270
Ala Ser Tyr Phe Ala Ala Ala Val Met Leu Pro Tyr Lys Ile Phe His
        275                 280                 285
Ser Glu Ala Glu Lys Ser Gly Tyr Asp Ile Glu Tyr Leu Gly Gln Leu
    290                 295                 300
Phe Gly Val Gly Tyr Glu Thr Thr Ala His Arg Leu Ser Thr Leu Gln
305                 310                 315                 320
Arg Pro Asn Leu Arg Gly Ile Pro Phe Thr Phe Val Arg Val Asp Arg
                325                 330                 335
Ala Gly Asn Met Ser Lys Arg Gln Ser Ala Thr Gly Phe His Phe Thr
            340                 345                 350
His Tyr Gly Gly Thr Cys Pro Leu Trp Asn Val Phe Glu Thr Phe Thr
        355                 360                 365
Asn Pro Gly Gln Val Leu Arg Gln Phe Ala Gln Met Pro Asp Gly Arg
    370                 375                 380
Asn Tyr Leu Trp Ile Ser Arg Thr Val Arg His His Glu Ala Arg Phe
385                 390                 395                 400
Gly Glu Val Asp Lys Met Phe Ala Ile Gly Leu Gly Cys Glu Ala Arg
                405                 410                 415
His Ala Asp Arg Thr Val Tyr Ser Arg Gly Phe Asn Leu Gln Asp Leu
            420                 425                 430
Ser Thr Ala Thr Pro Ile Gly Ser Gly Cys Arg Val Cys Thr Arg Glu
        435                 440                 445
Asn Cys Ala Gln Arg Ala Phe Pro Ser Val His Gly Arg Ile Asn Ile
    450                 455                 460
Asp Ala His Glu Ser Thr Ile Ala Pro Tyr
465                 470
```

<210> 38

<211> 18
<212> DNA
<213> Corynebacterium glutamicum

<400> 38
aaaaaaaaaa aaaaaaaa 18

<210> 39
<211> 17
<212> DNA
<213> Corynebacterium glutamicum

<400> 39
aaaaaaaaaa aaaaaaa 17

<210> 40
<211> 1386
<212> DNA
<213> Corynebacterium glutamicum

<220>
<221> CDS
<222> (1).. (1383)

<400> 40

```
atg tct gac aca ccg acc tca gct ctg atc acc acg gtc aac cgc agc      48
Met Ser Asp Thr Pro Thr Ser Ala Leu Ile Thr Thr Val Asn Arg Ser
1               5                  10                  15
ttc gat gga ttc gat ttg gaa gaa gta gca gca gac ctt gga gtt cgg      96
Phe Asp Gly Phe Asp Leu Glu Glu Val Ala Ala Asp Leu Gly Val Arg
            20                  25                  30
ctc acc tac ctg ccc gac gaa gaa cta gaa gta tcc aaa gtt ctc gcg     144
Leu Thr Tyr Leu Pro Asp Glu Glu Leu Glu Val Ser Lys Val Leu Ala
        35                  40                  45
gcg gac ctc ctc gct gag ggg cca gct ctc atc atc ggt gta gga aac     192
Ala Asp Leu Leu Ala Glu Gly Pro Ala Leu Ile Ile Gly Val Gly Asn
    50                  55                  60
acg ttt ttc gac gcc cag gtc gcc gct gcc ctc ggc gtc cca gtg cta     240
Thr Phe Phe Asp Ala Gln Val Ala Ala Ala Leu Gly Val Pro Val Leu
65                  70                  75                  80
```

```
ctg ctg gta gac aag caa ggc aag cac gtt gct ctt gct cgc acc cag      288
Leu Leu Val Asp Lys Gln Gly Lys His Val Ala Leu Ala Arg Thr Gln
                85                  90                  95

gta aac aat gcc ggc gca gtt gtt gca gca gca ttt acc gct gaa caa      336
Val Asn Asn Ala Gly Ala Val Val Ala Ala Ala Phe Thr Ala Glu Gln
            100                 105                 110

gag cca atg ccg gat aag ctg cgc aag gct gtg cgc aac cac agc aac      384
Glu Pro Met Pro Asp Lys Leu Arg Lys Ala Val Arg Asn His Ser Asn
        115                 120                 125

ctc gaa cca gtc atg agc gcc gaa ctc ttt gaa aac tgg ctg ctc aag      432
Leu Glu Pro Val Met Ser Ala Glu Leu Phe Glu Asn Trp Leu Leu Lys
    130                 135                 140

cgc gca cgc gca gag cac tcc cac att gtg ctg cca gaa ggt gac gac      480
Arg Ala Arg Ala Glu His Ser His Ile Val Leu Pro Glu Gly Asp Asp
145                 150                 155                 160

gac cgc atc ttg atg gct gcc cac cag ctg ctt gat caa gac atc tgt      528
Asp Arg Ile Leu Met Ala Ala His Gln Leu Leu Asp Gln Asp Ile Cys
                165                 170                 175

gac atc acg atc ctg ggc gat cca gta aag atc aag gag cgc gct acc      576
Asp Ile Thr Ile Leu Gly Asp Pro Val Lys Ile Lys Glu Arg Ala Thr
            180                 185                 190

gaa ctt ggc ctg cac ctt aac act gca tac ctg gtc aat ccg ctg aca      624
Glu Leu Gly Leu His Leu Asn Thr Ala Tyr Leu Val Asn Pro Leu Thr
        195                 200                 205

gat cct cgc ctg gag gaa ttc gcc gaa caa ttc gcg gag ctg cgc aag      672
Asp Pro Arg Leu Glu Glu Phe Ala Glu Gln Phe Ala Glu Leu Arg Lys
    210                 215                 220

tca aag agc gtc act atc gat gaa gcc cgc gaa atc atg aag gat att      720
Ser Lys Ser Val Thr Ile Asp Glu Ala Arg Glu Ile Met Lys Asp Ile
225                 230                 235                 240

tcc tac ttc ggc acc atg atg gtc cac aac ggc gac gcc gac gga atg      768
Ser Tyr Phe Gly Thr Met Met Val His Asn Gly Asp Ala Asp Gly Met
                245                 250                 255

gta tcc ggt gca gca aac acc acc gca cac acc att aag cca agc ttc      816
Val Ser Gly Ala Ala Asn Thr Thr Ala His Thr Ile Lys Pro Ser Phe
            260                 265                 270

cag atc atc aaa act gtt cca gaa gca tcc gtc gtt tct tcc atc ttc      864
Gln Ile Ile Lys Thr Val Pro Glu Ala Ser Val Val Ser Ser Ile Phe
        275                 280                 285

ctc atg gtg ctg cgc ggg cga ctg tgg gca ttc ggc gac tgt gct gtt      912
Leu Met Val Leu Arg Gly Arg Leu Trp Ala Phe Gly Asp Cys Ala Val
    290                 295                 300

aac ccg aac cca act gct gaa cag ctt ggt gaa atc gcc gtt gtg tca      960
Asn Pro Asn Pro Thr Ala Glu Gln Leu Gly Glu Ile Ala Val Val Ser
305                 310                 315                 320
```

```
gca aaa act gca gca caa ttt ggc att gat cct cgc gta gcc atc ttg      1008
Ala Lys Thr Ala Ala Gln Phe Gly Ile Asp Pro Arg Val Ala Ile Leu
                325                 330                 335
tcc tac tcc act ggc aac tcc ggc gga ggc tca gat gtg gat cgc gcc      1056
Ser Tyr Ser Thr Gly Asn Ser Gly Gly Gly Ser Asp Val Asp Arg Ala
            340                 345                 350
atc gac gct ctt gca gaa gca cgc cga ctt aac cca gaa cta tgc gtc      1104
Ile Asp Ala Leu Ala Glu Ala Arg Arg Leu Asn Pro Glu Leu Cys Val
        355                 360                 365
gat gga cca ctt cag ttc gac gcc gcc gtc gac ccg ggt gtg gcg cgc      1152
Asp Gly Pro Leu Gln Phe Asp Ala Ala Val Asp Pro Gly Val Ala Arg
    370                 375                 380
aag aag atg cca gac tct gac gtc gct ggc cag gca aat gtg ttt atc      1200
Lys Lys Met Pro Asp Ser Asp Val Ala Gly Gln Ala Asn Val Phe Ile
385                 390                 395                 400
ttc cct gac ctg gaa gcc gga aac atc ggc tac aaa act gca caa cgc      1248
Phe Pro Asp Leu Glu Ala Gly Asn Ile Gly Tyr Lys Thr Ala Gln Arg
                405                 410                 415
acc ggt cac gcc ctg gca gtt ggt ccg att ctg cag ggc cta aac aaa      1296
Thr Gly His Ala Leu Ala Val Gly Pro Ile Leu Gln Gly Leu Asn Lys
            420                 425                 430
cca gtc aac gac ctt tcc cgt ggc gca aca gtc cct gac atc gtc aac      1344
Pro Val Asn Asp Leu Ser Arg Gly Ala Thr Val Pro Asp Ile Val Asn
        435                 440                 445
aca gta gcc atc aca gca att cag gca gga gga cgc agc taa              1386
Thr Val Ala Ile Thr Ala Ile Gln Ala Gly Gly Arg Ser
    450                 455                 460
```

<210> 41
<211> 461
<212> PRT
<213> Corynebacterium glutamicum

<400> 41

```
Met Ser Asp Thr Pro Thr Ser Ala Leu Ile Thr Thr Val Asn Arg Ser
1               5                  10                  15
Phe Asp Gly Phe Asp Leu Glu Glu Val Ala Ala Asp Leu Gly Val Arg
            20                  25                  30
Leu Thr Tyr Leu Pro Asp Glu Glu Leu Glu Val Ser Lys Val Leu Ala
        35                  40                  45
Ala Asp Leu Leu Ala Glu Gly Pro Ala Leu Ile Ile Gly Val Gly Asn
    50                  55                  60
Thr Phe Phe Asp Ala Gln Val Ala Ala Ala Leu Gly Val Pro Val Leu
65                  70                  75                  80
```

Leu Leu Val Asp Lys Gln Gly Lys His Val Ala Leu Ala Arg Thr Gln
85 90 95
Val Asn Asn Ala Gly Ala Val Val Ala Ala Ala Phe Thr Ala Glu Gln
100 105 110
Glu Pro Met Pro Asp Lys Leu Arg Lys Ala Val Arg Asn His Ser Asn
115 120 125
Leu Glu Pro Val Met Ser Ala Glu Leu Phe Glu Asn Trp Leu Leu Lys
130 135 140
Arg Ala Arg Ala Glu His Ser His Ile Val Leu Pro Glu Gly Asp Asp
145 150 155 160
Asp Arg Ile Leu Met Ala Ala His Gln Leu Leu Asp Gln Asp Ile Cys
165 170 175
Asp Ile Thr Ile Leu Gly Asp Pro Val Lys Ile Lys Glu Arg Ala Thr
180 185 190
Glu Leu Gly Leu His Leu Asn Thr Ala Tyr Leu Val Asn Pro Leu Thr
195 200 205
Asp Pro Arg Leu Glu Glu Phe Ala Glu Gln Phe Ala Glu Leu Arg Lys
210 215 220
Ser Lys Ser Val Thr Ile Asp Glu Ala Arg Glu Ile Met Lys Asp Ile
225 230 235 240
Ser Tyr Phe Gly Thr Met Met Val His Asn Gly Asp Ala Asp Gly Met
245 250 255
Val Ser Gly Ala Ala Asn Thr Thr Ala His Thr Ile Lys Pro Ser Phe
260 265 270
Gln Ile Ile Lys Thr Val Pro Glu Ala Ser Val Val Ser Ser Ile Phe
275 280 285
Leu Met Val Leu Arg Gly Arg Leu Trp Ala Phe Gly Asp Cys Ala Val
290 295 300
Asn Pro Asn Pro Thr Ala Glu Gln Leu Gly Glu Ile Ala Val Val Ser
305 310 315 320
Ala Lys Thr Ala Ala Gln Phe Gly Ile Asp Pro Arg Val Ala Ile Leu
325 330 335
Ser Tyr Ser Thr Gly Asn Ser Gly Gly Gly Ser Asp Val Asp Arg Ala
340 345 350
Ile Asp Ala Leu Ala Glu Ala Arg Arg Leu Asn Pro Glu Leu Cys Val
355 360 365
Asp Gly Pro Leu Gln Phe Asp Ala Ala Val Asp Pro Gly Val Ala Arg
370 375 380
Lys Lys Met Pro Asp Ser Asp Val Ala Gly Gln Ala Asn Val Phe Ile
385 390 395 400
Phe Pro Asp Leu Glu Ala Gly Asn Ile Gly Tyr Lys Thr Ala Gln Arg
405 410 415
Thr Gly His Ala Leu Ala Val Gly Pro Ile Leu Gln Gly Leu Asn Lys
420 425 430
Pro Val Asn Asp Leu Ser Arg Gly Ala Thr Val Pro Asp Ile Val Asn

435 440 445
Thr Val Ala Ile Thr Ala Ile Gln Ala Gly Gly Arg Ser
450 455 460

<210> 42
<211> 1425
<212> DNA
<213> Corynebacterium glutamicum

<220>
<221> CDS
<222> (1)..(1922)

<400> 42

atg gga aag aca tat gtg ggg tcc agg ctg cgc caa ctg cgc cgc gaa 48
Met Gly Lys Thr Tyr Val Gly Ser Arg Leu Arg Gln Leu Arg Arg Glu
1 5 10 15
aga gac ctg agc cag gca tcc tta gca gca acc ctt ggc tta tct gca 96
Arg Asp Leu Ser Gln Ala Ser Leu Ala Ala Thr Leu Gly Leu Ser Ala
20 25 30
agt tat gta aat cag att gag cac gac gta cgc ccg ctc acc gta ccg 144
Ser Tyr Val Asn Gln Ile Glu His Asp Val Arg Pro Leu Thr Val Pro
35 40 45
gtg tta ttg cgc atc acc gag gcg ttc ggc gta gac gca acg ttt ttc 192
Val Leu Leu Arg Ile Thr Glu Ala Phe Gly Val Asp Ala Thr Phe Phe
50 55 60
tcc cgc gac gat gac tcc cgc ctg ctc gcc gag gtc caa gac gtc atg 240
Ser Arg Asp Asp Asp Ser Arg Leu Leu Ala Glu Val Gln Asp Val Met
65 70 75 80
ctg gac cgg gag atc aat cct gcg aac gtg gag ctg caa gag ctt tcg 288
Leu Asp Arg Glu Ile Asn Pro Ala Asn Val Glu Leu Gln Glu Leu Ser
85 90 95
gag atg gtg tac aac cac ccc caa cta gcg cgc gcg atg gtg gaa atg 336
Glu Met Val Tyr Asn His Pro Gln Leu Ala Arg Ala Met Val Glu Met
100 105 110
cac cag cgt tac cga aac gtg cgc gat aag ttc tcc atc gca gtg gat 384
His Gln Arg Tyr Arg Asn Val Arg Asp Lys Phe Ser Ile Ala Val Asp
115 120 125
aat cgc acc aac acg cct gag gaa cgc cgt ccc atc gcg gag gcc gtg 432
Asn Arg Thr Asn Thr Pro Glu Glu Arg Arg Pro Ile Ala Glu Ala Val
130 135 140
agc atg ccg cac gaa gag gtc cgc gat ttc att tac gcc cgc caa aac 480
Ser Met Pro His Glu Glu Val Arg Asp Phe Ile Tyr Ala Arg Gln Asn

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145                 150                 155                 160
tac ttc gat gcc ctt gac cgc cgc gcc gaa gcc atc gcc gcg caa ctg      528
Tyr Phe Asp Ala Leu Asp Arg Arg Ala Glu Ala Ile Ala Ala Gln Leu
                165                 170                 175
ggc tgg cag ccg tac gat tcc cgc gcc atg gaa gat tcg atc gcc cgc      576
Gly Trp Gln Pro Tyr Asp Ser Arg Ala Met Glu Asp Ser Ile Ala Arg
            180                 185                 190
cgc ctg caa atg gat cac gat gtc acc atc acc tcc tcc aaa gag gaa      624
Arg Leu Gln Met Asp His Asp Val Thr Ile Thr Ser Ser Lys Glu Glu
        195                 200                 205
tcc ggc acg ctg cac cac ttc gac ccc gag acg cgt ctg ctg aca atc      672
Ser Gly Thr Leu His His Phe Asp Pro Glu Thr Arg Leu Leu Thr Ile
    210                 215                 220
cac gca cgc ctc aac ccc ggg caa cgc gcc ttc cgc atg gcc acc gaa      720
His Ala Arg Leu Asn Pro Gly Gln Arg Ala Phe Arg Met Ala Thr Glu
225                 230                 235                 240
ctc ggc tac cta gaa gcc aac gac ctc atc gaa ggt atc gtt gac gac      768
Leu Gly Tyr Leu Glu Ala Asn Asp Leu Ile Glu Gly Ile Val Asp Asp
                245                 250                 255
ggc atc tgg tcc acc ccc gaa gcc cgc acc cta gcc atc cgc ggt gtg      816
Gly Ile Trp Ser Thr Pro Glu Ala Arg Thr Leu Ala Ile Arg Gly Val
            260                 265                 270
gcc tcc tac ttc gcc gcc gcc gtg atg ctg ccc tac aaa atc ttc cac      864
Ala Ser Tyr Phe Ala Ala Ala Val Met Leu Pro Tyr Lys Ile Phe His
        275                 280                 285
tcc gag gcc gaa aaa tcc ggc tac gac atc gag tac cta ggc caa ctc      912
Ser Glu Ala Glu Lys Ser Gly Tyr Asp Ile Glu Tyr Leu Gly Gln Leu
    290                 295                 300
ttt ggc gtg ggc tat gag aca acc gcc cac cgc ttg tcc acc ctg cag      960
Phe Gly Val Gly Tyr Glu Thr Thr Ala His Arg Leu Ser Thr Leu Gln
305                 310                 315                 320
cgc ccc aac ctg cgc ggc atc ccc ttt acc ttc gtg cgc gtc gac cgc     1008
Arg Pro Asn Leu Arg Gly Ile Pro Phe Thr Phe Val Arg Val Asp Arg
                325                 330                 335
gcc ggc aac atg tcc aaa cgc caa tcc gcc acc ggc ttc cac ttc acc     1056
Ala Gly Asn Met Ser Lys Arg Gln Ser Ala Thr Gly Phe His Phe Thr
            340                 345                 350
cac tac ggc ggc acc tgc ccc ctg tgg aac gtg ttt gaa acc ttc acc     1104
His Tyr Gly Gly Thr Cys Pro Leu Trp Asn Val Phe Glu Thr Phe Thr
        355                 360                 365
aac ccc ggc caa gtg ctc cgc caa ttc gcg caa atg ccc gac gga cgc     1152
Asn Pro Gly Gln Val Leu Arg Gln Phe Ala Gln Met Pro Asp Gly Arg
    370                 375                 380
aac tac ctg tgg atc tca cgc acc gtg cga cac cac gaa gcc cgg ttc     1200
Asn Tyr Leu Trp Ile Ser Arg Thr Val Arg His His Glu Ala Arg Phe
```

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385                 390                 395                 400
ggc gaa gta gac aaa atg ttc gcc atc ggc ctg ggc tgc gaa gcg cgc    1248
Gly Glu Val Asp Lys Met Phe Ala Ile Gly Leu Gly Cys Glu Ala Arg
                405                 410                 415
cac gcc gac cgc act gtg tac tcc cgc ggt ttc aac ctc cag gac ctc    1296
His Ala Asp Arg Thr Val Tyr Ser Arg Gly Phe Asn Leu Gln Asp Leu
            420                 425                 430
tcc acc gcc acc ccc atc ggg tcc ggc tgc cga gtg tgc acc cgc gag    1344
Ser Thr Ala Thr Pro Ile Gly Ser Gly Cys Arg Val Cys Thr Arg Glu
        435                 440                 445
aac tgc gcg cag cgc gca ttc cca tcc gtc cac ggc cgc atc aac atc    1392
Asn Cys Ala Gln Arg Ala Phe Pro Ser Val His Gly Arg Ile Asn Ile
    450                 455                 460
gac gcg cac gaa tcc act atc gcg ccg tac taa                       1425
Asp Ala His Glu Ser Thr Ile Ala Pro Tyr
465                 470
```

<210> 43
<211> 474
<212> PRT
<213> Corynebacterium glutamicum

<400> 43

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Met Gly Lys Thr Tyr Val Gly Ser Arg Leu Arg Gln Leu Arg Arg Glu
1               5                  10                  15
Arg Asp Leu Ser Gln Ala Ser Leu Ala Ala Thr Leu Gly Leu Ser Ala
            20                  25                  30
Ser Tyr Val Asn Gln Ile Glu His Asp Val Arg Pro Leu Thr Val Pro
        35                  40                  45
Val Leu Leu Arg Ile Thr Glu Ala Phe Gly Val Asp Ala Thr Phe Phe
    50                  55                  60
Ser Arg Asp Asp Asp Ser Arg Leu Leu Ala Glu Val Gln Asp Val Met
65                  70                  75                  80
Leu Asp Arg Glu Ile Asn Pro Ala Asn Val Glu Leu Gln Glu Leu Ser
                85                  90                  95
Glu Met Val Tyr Asn His Pro Gln Leu Ala Arg Ala Met Val Glu Met
            100                 105                 110
His Gln Arg Tyr Arg Asn Val Arg Asp Lys Phe Ser Ile Ala Val Asp
        115                 120                 125
Asn Arg Thr Asn Thr Pro Glu Glu Arg Arg Pro Ile Ala Glu Ala Val
    130                 135                 140
Ser Met Pro His Glu Glu Val Arg Asp Phe Ile Tyr Ala Arg Gln Asn
145                 150                 155                 160
Tyr Phe Asp Ala Leu Asp Arg Arg Ala Glu Ala Ile Ala Ala Gln Leu
```

165 170 175
Gly Trp Gln Pro Tyr Asp Ser Arg Ala Met Glu Asp Ser Ile Ala Arg
180 185 190
Arg Leu Gln Met Asp His Asp Val Thr Ile Thr Ser Ser Lys Glu Glu
195 200 205
Ser Gly Thr Leu His His Phe Asp Pro Glu Thr Arg Leu Leu Thr Ile
210 215 220
His Ala Arg Leu Asn Pro Gly Gln Arg Ala Phe Arg Met Ala Thr Glu
225 230 235 240
Leu Gly Tyr Leu Glu Ala Asn Asp Leu Ile Glu Gly Ile Val Asp Asp
245 250 255
Gly Ile Trp Ser Thr Pro Glu Ala Arg Thr Leu Ala Ile Arg Gly Val
260 265 270
Ala Ser Tyr Phe Ala Ala Ala Val Met Leu Pro Tyr Lys Ile Phe His
275 280 285
Ser Glu Ala Glu Lys Ser Gly Tyr Asp Ile Glu Tyr Leu Gly Gln Leu
290 295 300
Phe Gly Val Gly Tyr Glu Thr Thr Ala His Arg Leu Ser Thr Leu Gln
305 310 315 320
Arg Pro Asn Leu Arg Gly Ile Pro Phe Thr Phe Val Arg Val Asp Arg
325 330 335
Ala Gly Asn Met Ser Lys Arg Gln Ser Ala Thr Gly Phe His Phe Thr
340 345 350
His Tyr Gly Gly Thr Cys Pro Leu Trp Asn Val Phe Glu Thr Phe Thr
355 360 365
Asn Pro Gly Gln Val Leu Arg Gln Phe Ala Gln Met Pro Asp Gly Arg
370 375 380
Asn Tyr Leu Trp Ile Ser Arg Thr Val Arg His His Glu Ala Arg Phe
385 390 395 400
Gly Glu Val Asp Lys Met Phe Ala Ile Gly Leu Gly Cys Glu Ala Arg
405 410 415
His Ala Asp Arg Thr Val Tyr Ser Arg Gly Phe Asn Leu Gln Asp Leu
420 425 430
Ser Thr Ala Thr Pro Ile Gly Ser Gly Cys Arg Val Cys Thr Arg Glu
435 440 445
Asn Cys Ala Gln Arg Ala Phe Pro Ser Val His Gly Arg Ile Asn Ile
450 455 460
Asp Ala His Glu Ser Thr Ile Ala Pro Tyr
465 470

<210> 44
<211> 20
<212> DNA
<213> Artificial sequence

<220>
<223> primer

<400> 44 20
aatagagcgg gtcatacacc 20

<210> 45
<211> 44
<212> DNA
<213> Artificial sequence

<220>
<223> primer

<400> 45 44
ctctaaaaca ggaagagccc cttaaatagt gacagcaaga gcag 44

<210> 46
<211> 47
<212> DNA
<213> Artificial sequence

<220>
<223> primer

<400> 46
gcgctgctct tgctgtcact atttaagggg ctcttcctgt tttagag 47

<210> 47
C211> 40
<212> DNA
<213> Artificial sequence

<220>
<223> primer

<400> 47 40
ccacagtcat gaccttaaat ggcccttccg gttttggggt 40

<210> 48
<211> 40
<212> DNA
<213> Artificial sequence

<220>
<223> primer

<400> 48 40
accccaaaac cggaagggcc atttaaggtc atgactgtgg 40

<210> 49
<211> 20
<212> DNA
<213> Artificial sequence

<220>
<223> primer

<400> 49 20
gtggatgatt ttagaaccca 20

<210> 50
<211> 28
<212> DNA
<213> Artificial sequence

<220>
<223> primer

<400> 50 28
gggagctcga ctttctggct cctttact 28

<210> 51
<211> 28
<212> DNA
<213> Artificial sequence

<220>
<223> primer

<400> 51 28
gggagctcgc cgatgactaa taatgaga 28

<210> 52
<211> 2367
<212> DNA
<213> Lactobacillus pentosus

<220>
<221> CDS
<222> (1)..(2367)
<223>

(400)

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atg tct aca gat tac tca tca cca gca tat ttg caa aaa gtt gat aag      48
Met Ser Thr Asp Tyr Ser Ser Pro Ala Tyr Leu Gln Lys Val Asp Lys
1               5                  10                  15

tac tgg cgt gct gcc aac tat tta tca gtt ggt caa ctt tat tta aaa      96
Tyr Trp Arg Ala Ala Asn Tyr Leu Ser Val Gly Gln Leu Tyr Leu Lys
            20                  25                  30

gat aat cct tta tta caa cgg cca tta aag gct agt gac gtt aag gtt     144
Asp Asn Pro Leu Leu Gln Arg Pro Leu Lys Ala Ser Asp Val Lys Val
        35                  40                  45

cac cca atc ggt cac tgg ggc acg att gcc ggc caa aac ttc atc tat     192
His Pro Ile Gly His Trp Gly Thr Ile Ala Gly Gln Asn Phe Ile Tyr
    50                  55                  60

gcg cat ctt aac cgg gtc atc aac aag tac ggt ttg aag atg ttc tac     240
Ala His Leu Asn Arg Val Ile Asn Lys Tyr Gly Leu Lys Met Phe Tyr
65                  70                  75                  80

gtt gaa ggt cca ggt cat ggt ggc caa gtg atg gtc tcc aac tca tac     288
Val Glu Gly Pro Gly His Gly Gly Gln Val Met Val Ser Asn Ser Tyr
                85                  90                  95

ctt gat ggg act tac acg gat att tat cct gaa att acg cag gat gtt     336
Leu Asp Gly Thr Tyr Thr Asp Ile Tyr Pro Glu Ile Thr Gln Asp Val
            100                 105                 110

gaa ggg atg caa aaa ctc ttc aag caa ttc tca ttc cca ggt ggc gtg     384
Glu Gly Met Gln Lys Leu Phe Lys Gln Phe Ser Phe Pro Gly Gly Val
        115                 120                 125

gct tcc cat gct gct cct gaa aca cca ggc tca atc cac gaa ggt ggc     432
Ala Ser His Ala Ala Pro Glu Thr Pro Gly Ser Ile His Glu Gly Gly
    130                 135                 140

gaa ctt ggt tac tca att tca cac ggt gtt ggg gca atc ctt gac aac     480
Glu Leu Gly Tyr Ser Ile Ser His Gly Val Gly Ala Ile Leu Asp Asn
145                 150                 155                 160

cct gat gaa atc gcc gca gtc gtt gtt ggt gat ggg gaa tcc gaa acc     528
Pro Asp Glu Ile Ala Ala Val Val Val Gly Asp Gly Glu Ser Glu Thr
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                    165                 170                 175
ggc cca tta gca act tca tgg caa tca acg aag ttc atc aac cca atc      576
Gly Pro Leu Ala Thr Ser Trp Gln Ser Thr Lys Phe Ile Asn Pro Ile
                180                 185                 190
aac gat ggg gca gtg tta cca atc ttg aac ctt aac ggc ttt aag att      624
Asn Asp Gly Ala Val Leu Pro Ile Leu Asn Leu Asn Gly Phe Lys Ile
        195                 200                 205
tct aac cca acg att ttt ggt cgg act tct gat gaa aag atc aag caa      672
Ser Asn Pro Thr Ile Phe Gly Arg Thr Ser Asp Glu Lys Ile Lys Gln
    210                 215                 220
tac ttc gaa agc atg aac tgg gaa cca atc ttt gtt gaa ggt gac gat      720
Tyr Phe Glu Ser Met Asn Trp Glu Pro Ile Phe Val Glu Gly Asp Asp
225                 230                 235                 240
cct gaa aag gtt cac cca gct tta gct aag gcc atg gat gaa gcc gtc      768
Pro Glu Lys Val His Pro Ala Leu Ala Lys Ala Met Asp Glu Ala Val
                245                 250                 255
gaa aag atc aaa gcc att caa aag aac gct cgt gaa aac gat gac gct      816
Glu Lys Ile Lys Ala Ile Gln Lys Asn Ala Arg Glu Asn Asp Asp Ala
            260                 265                 270
act tta cca gta tgg ccg atg atc gtc ttc cgc gca cct aag ggc tgg      864
Thr Leu Pro Val Trp Pro Met Ile Val Phe Arg Ala Pro Lys Gly Trp
        275                 280                 285
act ggt cct aag tca tgg gat ggc gac aag atc gaa ggt tca ttc cga      912
Thr Gly Pro Lys Ser Trp Asp Gly Asp Lys Ile Glu Gly Ser Phe Arg
    290                 295                 300
gct cac caa att cca att cct gtt gac caa acc gac atg gaa cat gcc      960
Ala His Gln Ile Pro Ile Pro Val Asp Gln Thr Asp Met Glu His Ala
305                 310                 315                 320
gat gcg tta gtt gac tgg ttg gaa tca tat caa cca aag gaa ctc ttc     1008
Asp Ala Leu Val Asp Trp Leu Glu Ser Tyr Gln Pro Lys Glu Leu Phe
                325                 330                 335
aat gaa gat ggt tct ttg aag gat gat atc aaa gaa att atc cca act     1056
Asn Glu Asp Gly Ser Leu Lys Asp Asp Ile Lys Glu Ile Ile Pro Thr
            340                 345                 350
ggc gat gca cgg atg gcc gct aac cca atc act aat ggt ggg gtt gat     1104
Gly Asp Ala Arg Met Ala Ala Asn Pro Ile Thr Asn Gly Gly Val Asp
        355                 360                 365
cca aag gcc ttg aac tta cct aac ttc cgt gat tac gcc gtt gat acg     1152
Pro Lys Ala Leu Asn Leu Pro Asn Phe Arg Asp Tyr Ala Val Asp Thr
    370                 375                 380
tct aag cat ggt gcc aac gtt aag caa gat atg atc gtt tgg tca gac     1200
Ser Lys His Gly Ala Asn Val Lys Gln Asp Met Ile Val Trp Ser Asp
385                 390                 395                 400
tac ttg cgt gat gtt atc aag aag aac cca gat aac ttc cgg tta ttt     1248
Tyr Leu Arg Asp Val Ile Lys Lys Asn Pro Asp Asn Phe Arg Leu Phe
```

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                405                 410                 415
ggc cct gat gaa acc atg tca aac cgg tta tat ggt gtc ttt gaa acc   1296
Gly Pro Asp Glu Thr Met Ser Asn Arg Leu Tyr Gly Val Phe Glu Thr
            420                 425                 430
act aac cgt caa tgg atg gaa gat att cac cca gat agt gac caa tac   1344
Thr Asn Arg Gln Trp Met Glu Asp Ile His Pro Asp Ser Asp Gln Tyr
        435                 440                 445
gaa gca cct gct ggc cgg gtc ttg gat gct caa tta tct gaa cac caa   1392
Glu Ala Pro Ala Gly Arg Val Leu Asp Ala Gln Leu Ser Glu His Gln
    450                 455                 460
gct gaa ggt tgg tta gaa ggt tac gtc tta act ggt cgt cat ggc ttg   1440
Ala Glu Gly Trp Leu Glu Gly Tyr Val Leu Thr Gly Arg His Gly Leu
465                 470                 475                 480
ttt gca agt tac gaa gcc ttc tta cgg gtt gtc gac tca atg ttg acg   1488
Phe Ala Ser Tyr Glu Ala Phe Leu Arg Val Val Asp Ser Met Leu Thr
                485                 490                 495
caa cac ttc aag tgg tta cgt aag gcc aac gaa ctt gac tgg cgg aag   1536
Gln His Phe Lys Trp Leu Arg Lys Ala Asn Glu Leu Asp Trp Arg Lys
            500                 505                 510
aag tac ccg tca ctc aac att atc gcg gct tca act gtg ttc caa caa   1584
Lys Tyr Pro Ser Leu Asn Ile Ile Ala Ala Ser Thr Val Phe Gln Gln
        515                 520                 525
gac cat aat ggg tac acc cac caa gat cca ggt gcc ttg act cat ttg   1632
Asp His Asn Gly Tyr Thr His Gln Asp Pro Gly Ala Leu Thr His Leu
    530                 535                 540
gct gaa aag aag cct gaa tat atc cgc gaa tat tta cca gcc gac gcc   1680
Ala Glu Lys Lys Pro Glu Tyr Ile Arg Glu Tyr Leu Pro Ala Asp Ala
545                 550                 555                 560
aac tcc ttg tta gct gtt ggg gac gtc atc ttc cgt agc caa gaa aag   1728
Asn Ser Leu Leu Ala Val Gly Asp Val Ile Phe Arg Ser Gln Glu Lys
                565                 570                 575
atc aac tac gtg gtt acg tcg aag cac cca cgt caa caa tgg ttc agc   1776
Ile Asn Tyr Val Val Thr Ser Lys His Pro Arg Gln Gln Trp Phe Ser
            580                 585                 590
att gaa gaa gct aag caa tta gtt gac aac ggt ctt ggt atc att gac   1824
Ile Glu Glu Ala Lys Gln Leu Val Asp Asn Gly Leu Gly Ile Ile Asp
        595                 600                 605
tgg gca agc acg gac caa ggt agc gaa cca gat atc gtg ttt gct gct   1872
Trp Ala Ser Thr Asp Gln Gly Ser Glu Pro Asp Ile Val Phe Ala Ala
    610                 615                 620
gcc gga acg gaa cca acg ctt gaa acg ttg gct gca atc caa ttg ctc   1920
Ala Gly Thr Glu Pro Thr Leu Glu Thr Leu Ala Ala Ile Gln Leu Leu
625                 630                 635                 640
cat gat agc ttc cca gac atg aag att cgt ttc gtg aac gtg gtc gac   1968
His Asp Ser Phe Pro Asp Met Lys Ile Arg Phe Val Asn Val Val Asp
```

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                645                 650                 655
atc ttg aag tta cgt agc cct gaa aag gac cct cgt ggc ttg tca gat    2016
Ile Leu Lys Leu Arg Ser Pro Glu Lys Asp Pro Arg Gly Leu Ser Asp
            660                 665                 670
gct gaa ttt gac cat tac ttc act aag gac aaa cca gtt gtc ttc gcc    2064
Ala Glu Phe Asp His Tyr Phe Thr Lys Asp Lys Pro Val Val Phe Ala
        675                 680                 685
ttc cat ggt tac gaa gac ctg gtt cgt gac atc ttc ttt gat cgt cac    2112
Phe His Gly Tyr Glu Asp Leu Val Arg Asp Ile Phe Phe Asp Arg His
    690                 695                 700
aac cac aac tta cac gtg cat ggc tac cgt gaa aat ggt gac att acg    2160
Asn His Asn Leu His Val His Gly Tyr Arg Glu Asn Gly Asp Ile Thr
705                 710                 715                 720
aca cca ttc gat gtc cgg gtc atg aac caa atg gac cgt ttc gac tta    2208
Thr Pro Phe Asp Val Arg Val Met Asn Gln Met Asp Arg Phe Asp Leu
                725                 730                 735
gca aaa tct gca att gcg gcg caa cca gca atg gaa aac acc ggt gca    2256
Ala Lys Ser Ala Ile Ala Ala Gln Pro Ala Met Glu Asn Thr Gly Ala
            740                 745                 750
gcc ttt gtt caa gac atg gat aac atg ctt gca aaa cac aac gca tac    2304
Ala Phe Val Gln Asp Met Asp Asn Met Leu Ala Lys His Asn Ala Tyr
        755                 760                 765
atc cgt gac gcc gga acc gac ttg cca gaa gtt aac gac tgg caa tgg    2352
Ile Arg Asp Ala Gly Thr Asp Leu Pro Glu Val Asn Asp Trp Gln Trp
    770                 775                 780
aaa ggt ttg aaa taa                                               2367
Lys Gly Leu Lys
785
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<210> 53
<211> 788
<212> PRT
<213> Lactobacillus pentosus

<400> 53

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Met Ser Thr Asp Tyr Ser Ser Pro Ala Tyr Leu Gln Lys Val Asp Lys
1               5                  10                  15
Tyr Trp Arg Ala Ala Asn Tyr Leu Ser Val Gly Gln Leu Tyr Leu Lys
            20                  25                  30
Asp Asn Pro Leu Leu Gln Arg Pro Leu Lys Ala Ser Asp Val Lys Val
        35                  40                  45
His Pro Ile Gly His Trp Gly Thr Ile Ala Gly Gln Asn Phe Ile Tyr
    50                  55                  60
```

Ala His Leu Asn Arg Val Ile Asn Lys Tyr Gly Leu Lys Met Phe Tyr
65 70 75 80
Val Glu Gly Pro Gly His Gly Gly Gln Val Met Val Ser Asn Ser Tyr
85 90 95
Leu Asp Gly Thr Tyr Thr Asp Ile Tyr Pro Glu Ile Thr Gln Asp Val
100 105 110
Glu Gly Met Gln Lys Leu Phe Lys Gln Phe Ser Phe Pro Gly Gly Val
115 120 125
Ala Ser His Ala Ala Pro Glu Thr Pro Gly Ser Ile His Glu Gly Gly
130 135 140
Glu Leu Gly Tyr Ser Ile Ser His Gly Val Gly Ala Ile Leu Asp Asn
145 150 155 160
Pro Asp Glu Ile Ala Ala Val Val Val Gly Asp Gly Glu Ser Glu Thr
165 170 175

Gly Pro Leu Ala Thr Ser Trp Gln Ser Thr Lys Phe Ile Asn Pro Ile
180 185 190
Asn Asp Gly Ala Val Leu Pro Ile Leu Asn Leu Asn Gly Phe Lys Ile
195 200 205
Ser Asn Pro Thr Ile Phe Gly Arg Thr Ser Asp Glu Lys Ile Lys Gln
210 215 220
Tyr Phe Glu Ser Met Asn Trp Glu Pro Ile Phe Val Glu Gly Asp Asp
225 230 235 240
Pro Glu Lys Val His Pro Ala Leu Ala Lys Ala Met Asp Glu Ala Val
245 250 255
Glu Lys Ile Lys Ala Ile Gln Lys Asn Ala Arg Glu Asn Asp Asp Ala
260 265 270
Thr Leu Pro Val Trp Pro Met Ile Val Phe Arg Ala Pro Lys Gly Trp
275 280 285
Thr Gly Pro Lys Ser Trp Asp Gly Asp Lys Ile Glu Gly Ser Phe Arg
290 295 300
Ala His Gln Ile Pro Ile Pro Val Asp Gln Thr Asp Met Glu His Ala
305 310 315 320
Asp Ala Leu Val Asp Trp Leu Glu Ser Tyr Gln Pro Lys Glu Leu Phe
325 330 335
Asn Glu Asp Gly Ser Leu Lys Asp Asp Ile Lys Glu Ile Ile Pro Thr
340 345 350
Gly Asp Ala Arg Met Ala Ala Asn Pro Ile Thr Asn Gly Gly Val Asp
355 360 365
Pro Lys Ala Leu Asn Leu Pro Asn Phe Arg Asp Tyr Ala Val Asp Thr
370 375 380
Ser Lys His Gly Ala Asn Val Lys Gln Asp Met Ile Val Trp Ser Asp
385 390 395 400
Tyr Leu Arg Asp Val Ile Lys Lys Asn Pro Asp Asn Phe Arg Leu Phe
405 410 415

Gly Pro Asp Glu Thr Met Ser Asn Arg Leu Tyr Gly Val Phe Glu Thr
420 425 430
Thr Asn Arg Gln Trp Met Glu Asp Ile His Pro Asp Ser Asp Gln Tyr
435 440 445
Glu Ala Pro Ala Gly Arg Val Leu Asp Ala Gln Leu Ser Glu His Gln
450 455 460
Ala Glu Gly Trp Leu Glu Gly Tyr Val Leu Thr Gly Arg His Gly Leu
465 470 475 480
Phe Ala Ser Tyr Glu Ala Phe Leu Arg Val Val Asp Ser Met Leu Thr
485 490 495
Gln His Phe Lys Trp Leu Arg Lys Ala Asn Glu Leu Asp Trp Arg Lys
500 505 510
Lys Tyr Pro Ser Leu Asn Ile Ile Ala Ala Ser Thr Val Phe Gln Gln
515 520 525
Asp His Asn Gly Tyr Thr His Gln Asp Pro Gly Ala Leu Thr His Leu
530 535 540
Ala Glu Lys Lys Pro Glu Tyr Ile Arg Glu Tyr Leu Pro Ala Asp Ala
545 550 555 560
Asn Ser Leu Leu Ala Val Gly Asp Val Ile Phe Arg Ser Gln Glu Lys
565 570 575
Ile Asn Tyr Val Val Thr Ser Lys His Pro Arg Gln Gln Trp Phe Ser
580 585 590
Ile Glu Glu Ala Lys Gln Leu Val Asp Asn Gly Leu Gly Ile Ile Asp
595 600 605
Trp Ala Ser Thr Asp Gln Gly Ser Glu Pro Asp Ile Val Phe Ala Ala
610 615 620
Ala Gly Thr Glu Pro Thr Leu Glu Thr Leu Ala Ala Ile Gln Leu Leu
625 630 635 640
His Asp Ser Phe Pro Asp Met Lys Ile Arg Phe Val Asn Val Val Asp
645 650 655
Ile Leu Lys Leu Arg Ser Pro Glu Lys Asp Pro Arg Gly Leu Ser Asp
660 665 670
Ala Glu Phe Asp His Tyr Phe Thr Lys Asp Lys Pro Val Val Phe Ala
675 680 685
Phe His Gly Tyr Glu Asp Leu Val Arg Asp Ile Phe Phe Asp Arg His
690 695 700
Asn His Asn Leu His Val His Gly Tyr Arg Glu Asn Gly Asp Ile Thr
705 710 715 720
Thr Pro Phe Asp Val Arg Val Met Asn Gln Met Asp Arg Phe Asp Leu
725 730 735
Ala Lys Ser Ala Ile Ala Ala Gln Pro Ala Met Glu Asn Thr Gly Ala
740 745 750
Ala Phe Val Gln Asp Met Asp Asn Met Leu Ala Lys His Asn Ala Tyr
755 760 765
Ile Arg Asp Ala Gly Thr Asp Leu Pro Glu Val Asn Asp Trp Gln Trp

770 775 780

Lys Gly Leu Lys
785

<210> 54
<211> 2367
<212> DNA
<213> Lactobacillus plantarum

<220>
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<222> (1).. (2367)
<223>

<400> 54

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atg aca aca gat tac tca tca cca gca tat ttg caa aaa gtt gat aag      48
Met Thr Thr Asp Tyr Ser Ser Pro Ala Tyr Leu Gln Lys Val Asp Lys
1               5                  10                  15
tac tgg cgt gct gcc aac tac tta tca gtt ggt caa ctt tat tta aaa      96
Tyr Trp Arg Ala Ala Asn Tyr Leu Ser Val Gly Gln Leu Tyr Leu Lys
            20                  25                  30
gat aat cca cta tta caa cgg cca ttg aag gcc agt gac gtt aag gtt     144
Asp Asn Pro Leu Leu Gln Arg Pro Leu Lys Ala Ser Asp Val Lys Val
        35                  40                  45
cat cca att ggt cac tgg ggg acg att gcc ggt caa aac ttt atc tat     192
His Pro Ile Gly His Trp Gly Thr Ile Ala Gly Gln Asn Phe Ile Tyr
    50                  55                  60
gct cat ctt aac cgg gtc atc aac aag tac ggt ttg aag atg ttc tac     240
Ala His Leu Asn Arg Val Ile Asn Lys Tyr Gly Leu Lys Met Phe Tyr
65                  70                  75                  80
gtt gaa ggt cca ggt cat ggt ggt caa gtg atg gtt tca aac tct tac     288
Val Glu Gly Pro Gly His Gly Gly Gln Val Met Val Ser Asn Ser Tyr
                85                  90                  95
ctt gac ggt act tac acc gat att tat cca gaa att acg cag gat gtt     336
Leu Asp Gly Thr Tyr Thr Asp Ile Tyr Pro Glu Ile Thr Gln Asp Val
            100                 105                 110
gaa ggg atg caa aag ctc ttc aag caa ttc tca ttc cca ggt ggg gtt     384
Glu Gly Met Gln Lys Leu Phe Lys Gln Phe Ser Phe Pro Gly Gly Val
        115                 120                 125
gct tcc cat gcg gca cct gaa aca ccc ggt tca atc cac gaa ggt ggc     432
Ala Ser His Ala Ala Pro Glu Thr Pro Gly Ser Ile His Glu Gly Gly
    130                 135                 140
gaa ctt ggt tac tca att tca cac ggg gtt ggg gca att ctt gac aat     480
Glu Leu Gly Tyr Ser Ile Ser His Gly Val Gly Ala Ile Leu Asp Asn
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145                 150                 155                 160
cct gac gaa atc gcc gcg gtt gtt gtt ggt gat ggg gaa tcc gaa acg      528
Pro Asp Glu Ile Ala Ala Val Val Val Gly Asp Gly Glu Ser Glu Thr
                165                 170                 175
ggt cca tta gca act tca tgg caa tca acg aag ttc att aac cca atc      576
Gly Pro Leu Ala Thr Ser Trp Gln Ser Thr Lys Phe Ile Asn Pro Ile
            180                 185                 190
aac gac ggg gct gtt tta cca atc ttg aac tta aat ggt ttt aag att      624
Asn Asp Gly Ala Val Leu Pro Ile Leu Asn Leu Asn Gly Phe Lys Ile
        195                 200                 205
tct aat cca acg att ttt ggt cgg act tct gat gct aag att aag gaa      672
Ser Asn Pro Thr Ile Phe Gly Arg Thr Ser Asp Ala Lys Ile Lys Glu
    210                 215                 220
tac ttc gaa agc atg aat tgg gaa cca atc ttc gtt gaa ggt gac gat      720
Tyr Phe Glu Ser Met Asn Trp Glu Pro Ile Phe Val Glu Gly Asp Asp
225                 230                 235                 240
cct gaa aag gtt cac cca gcc tta gct aag gcc atg gat gaa gcc gtt      768
Pro Glu Lys Val His Pro Ala Leu Ala Lys Ala Met Asp Glu Ala Val
                245                 250                 255
gaa aag atc aag gca atc cag aag cat gct cgc gaa aat aac gat gca      816
Glu Lys Ile Lys Ala Ile Gln Lys His Ala Arg Glu Asn Asn Asp Ala
            260                 265                 270
aca ttg cca gta tgg cca atg atc gtc ttc cgc gca cct aag ggc tgg      864
Thr Leu Pro Val Trp Pro Met Ile Val Phe Arg Ala Pro Lys Gly Trp
        275                 280                 285
act ggt ccg aag tca tgg gac ggt gat aag atc gaa ggt tca ttc cgt      912
Thr Gly Pro Lys Ser Trp Asp Gly Asp Lys Ile Glu Gly Ser Phe Arg
    290                 295                 300
gct cat caa att ccg att cct gtt gat caa aat gac atg gaa cat gcg      960
Ala His Gln Ile Pro Ile Pro Val Asp Gln Asn Asp Met Glu His Ala
305                 310                 315                 320
gat gct tta gtt gat tgg ctc gaa tca tat caa cca aaa gaa ctc ttc     1008
Asp Ala Leu Val Asp Trp Leu Glu Ser Tyr Gln Pro Lys Glu Leu Phe
                325                 330                 335
aat gaa gat ggc tct ttg aag gat gat att aaa gaa att att cct act     1056
Asn Glu Asp Gly Ser Leu Lys Asp Asp Ile Lys Glu Ile Ile Pro Thr
            340                 345                 350
ggg gac agt cgg atg gct gct aac cca atc acc aat ggt ggg gtc gat     1104
Gly Asp Ser Arg Met Ala Ala Asn Pro Ile Thr Asn Gly Gly Val Asp
        355                 360                 365
ccg aaa gcc ttg aac tta cca aac ttc cgt gat tat gcg gtc gat acg     1152
Pro Lys Ala Leu Asn Leu Pro Asn Phe Arg Asp Tyr Ala Val Asp Thr
    370                 375                 380
tcc aaa gaa ggc gcg aat gtt aag caa gat atg atc gtt tgg tca gac     1200
Ser Lys Glu Gly Ala Asn Val Lys Gln Asp Met Ile Val Trp Ser Asp
```

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385                 390                 395                 400
tat ttg cgg gat gtc atc aag aaa aat cct gat aac ttc cgg ttg ttc   1248
Tyr Leu Arg Asp Val Ile Lys Lys Asn Pro Asp Asn Phe Arg Leu Phe
                405                 410                 415
gga cct gat gaa acc atg tct aac cgt tta tat ggt gtc ttc gaa acc   1296
Gly Pro Asp Glu Thr Met Ser Asn Arg Leu Tyr Gly Val Phe Glu Thr
            420                 425                 430
act aat cgt caa tgg atg gaa gac att cat cca gat agt gac caa tat   1344
Thr Asn Arg Gln Trp Met Glu Asp Ile His Pro Asp Ser Asp Gln Tyr
        435                 440                 445
gaa gca cca gct ggc cgg gtc tta gat gct cag tta tct gaa cac caa   1392
Glu Ala Pro Ala Gly Arg Val Leu Asp Ala Gln Leu Ser Glu His Gln
    450                 455                 460
gct gaa ggt tgg tta gaa ggt tac gtc tta act gga cgt cat ggg tta   1440
Ala Glu Gly Trp Leu Glu Gly Tyr Val Leu Thr Gly Arg His Gly Leu
465                 470                 475                 480
ttt gcc agt tat gaa gcc ttc cta cgc gtt gtg gac tca atg ttg acg   1488
Phe Ala Ser Tyr Glu Ala Phe Leu Arg Val Val Asp Ser Met Leu Thr
                485                 490                 495
caa cac ttc aag tgg tta cgt aaa gcc aat gaa ctt gat tgg cgt aaa   1536
Gln His Phe Lys Trp Leu Arg Lys Ala Asn Glu Leu Asp Trp Arg Lys
            500                 505                 510
aag tac cca tca ctt aac att atc gcg gct tca act gta ttc caa caa   1584
Lys Tyr Pro Ser Leu Asn Ile Ile Ala Ala Ser Thr Val Phe Gln Gln
        515                 520                 525
gac cat aat ggt tat acc cac caa gat cca ggt gca tta act cat ttg   1632
Asp His Asn Gly Tyr Thr His Gln Asp Pro Gly Ala Leu Thr His Leu
    530                 535                 540
gcc gaa aag aaa cca gaa tac att cgt gaa tat tta cca gcc gat gcc   1680
Ala Glu Lys Lys Pro Glu Tyr Ile Arg Glu Tyr Leu Pro Ala Asp Ala
545                 550                 555                 560
aac acg tta tta gct gtc ggt gac gtc att ttc cgg agc caa gaa aag   1728
Asn Thr Leu Leu Ala Val Gly Asp Val Ile Phe Arg Ser Gln Glu Lys
                565                 570                 575
atc aac tac gtg gtt acg tca aaa cac cca cgt caa caa tgg ttc agc   1776
Ile Asn Tyr Val Val Thr Ser Lys His Pro Arg Gln Gln Trp Phe Ser
            580                 585                 590
att gaa gaa gct aag caa tta gtt gac aat ggt ctt ggt atc att gat   1824
Ile Glu Glu Ala Lys Gln Leu Val Asp Asn Gly Leu Gly Ile Ile Asp
        595                 600                 605
tgg gca agt acg gac caa ggt agc gaa cca gac att gtc ttt gca gct   1872
Trp Ala Ser Thr Asp Gln Gly Ser Glu Pro Asp Ile Val Phe Ala Ala
    610                 615                 620
gct ggg acg gaa cca acg ctt gaa acg ttg gct gcc atc caa tta cta   1920
Ala Gly Thr Glu Pro Thr Leu Glu Thr Leu Ala Ala Ile Gln Leu Leu
```

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625                 630                 635                 640
cac gac agt ttc cca gag atg aag att cgt ttc gtg aac gtg gtc gac  1968
His Asp Ser Phe Pro Glu Met Lys Ile Arg Phe Val Asn Val Val Asp
                645                 650                 655
atc ttg aag tta cgt agt cct gaa aag gat ccg cgg ggc ttg tca gat  2016
Ile Leu Lys Leu Arg Ser Pro Glu Lys Asp Pro Arg Gly Leu Ser Asp
            660                 665                 670
gct gag ttt gac cat tac ttt act aag gac aaa cca gtg gtc ttt gct  2064
Ala Glu Phe Asp His Tyr Phe Thr Lys Asp Lys Pro Val Val Phe Ala
        675                 680                 685
ttc cac ggt tac gaa gac tta gtt cgt gac atc ttc ttt gat cgt cac  2112
Phe His Gly Tyr Glu Asp Leu Val Arg Asp Ile Phe Phe Asp Arg His
    690                 695                 700
aac cat aac tta tac gtc cac ggt tac cgt gaa aat ggt gat att acc  2160
Asn His Asn Leu Tyr Val His Gly Tyr Arg Glu Asn Gly Asp Ile Thr
705                 710                 715                 720
aca cca ttc gac gta cgg gtc atg aac cag atg gac cgc ttc gac tta  2208
Thr Pro Phe Asp Val Arg Val Met Asn Gln Met Asp Arg Phe Asp Leu
                725                 730                 735
gct aag tcg gca att gcg gcg caa cca gca atg gaa aac act ggt gcg  2256
Ala Lys Ser Ala Ile Ala Ala Gln Pro Ala Met Glu Asn Thr Gly Ala
            740                 745                 750
gcc ttc gtt caa tcc atg gat aat atg ctt gct aaa cac aat gcc tat  2304
Ala Phe Val Gln Ser Met Asp Asn Met Leu Ala Lys His Asn Ala Tyr
        755                 760                 765
atc cgg gat gcc gga act gac ttg cca gaa gtt aat gat tgg caa tgg  2352
Ile Arg Asp Ala Gly Thr Asp Leu Pro Glu Val Asn Asp Trp Gln Trp
    770                 775                 780
aag ggt tta aaa taa                                             2367
Lys Gly Leu Lys
785
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<210> 55
<211> 788
<212> PRT
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<400> 55

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1               5                  10                  15
Tyr Trp Arg Ala Ala Asn Tyr Leu Ser Val Gly Gln Leu Tyr Leu Lys
            20                  25                  30
Asp Asn Pro Leu Leu Gln Arg Pro Leu Lys Ala Ser Asp Val Lys Val
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          35                  40                  45
His Pro Ile Gly His Trp Gly Thr Ile Ala Gly Gln Asn Phe Ile Tyr
    50                  55                  60
Ala His Leu Asn Arg Val Ile Asn Lys Tyr Gly Leu Lys Met Phe Tyr
65                  70                  75                  80
Val Glu Gly Pro Gly His Gly Gly Gln Val Met Val Ser Asn Ser Tyr
                85                  90                  95
Leu Asp Gly Thr Tyr Thr Asp Ile Tyr Pro Glu Ile Thr Gln Asp Val
            100                 105                 110
Glu Gly Met Gln Lys Leu Phe Lys Gln Phe Ser Phe Pro Gly Gly Val
        115                 120                 125
Ala Ser His Ala Ala Pro Glu Thr Pro Gly Ser Ile His Glu Gly Gly
    130                 135                 140
Glu Leu Gly Tyr Ser Ile Ser His Gly Val Gly Ala Ile Leu Asp Asn
145                 150                 155                 160
Pro Asp Glu Ile Ala Ala Val Val Val Gly Asp Gly Glu Ser Glu Thr
                165                 170                 175
Gly Pro Leu Ala Thr Ser Trp Gln Ser Thr Lys Phe Ile Asn Pro Ile
            180                 185                 190
Asn Asp Gly Ala Val Leu Pro Ile Leu Asn Leu Asn Gly Phe Lys Ile
        195                 200                 205
Ser Asn Pro Thr Ile Phe Gly Arg Thr Ser Asp Ala Lys Ile Lys Glu
    210                 215                 220
Tyr Phe Glu Ser Met Asn Trp Glu Pro Ile Phe Val Glu Gly Asp Asp
225                 230                 235                 240
Pro Glu Lys Val His Pro Ala Leu Ala Lys Ala Met Asp Glu Ala Val
                245                 250                 255
Glu Lys Ile Lys Ala Ile Gln Lys His Ala Arg Glu Asn Asn Asp Ala
            260                 265                 270
Thr Leu Pro Val Trp Pro Met Ile Val Phe Arg Ala Pro Lys Gly Trp
        275                 280                 285
Thr Gly Pro Lys Ser Trp Asp Gly Asp Lys Ile Glu Gly Ser Phe Arg
    290                 295                 300
Ala His Gln Ile Pro Ile Pro Val Asp Gln Asn Asp Met Glu His Ala
305                 310                 315                 320
Asp Ala Leu Val Asp Trp Leu Glu Ser Tyr Gln Pro Lys Glu Leu Phe
                325                 330                 335
Asn Glu Asp Gly Ser Leu Lys Asp Asp Ile Lys Glu Ile Ile Pro Thr
            340                 345                 350
Gly Asp Ser Arg Met Ala Ala Asn Pro Ile Thr Asn Gly Gly Val Asp
        355                 360                 365
Pro Lys Ala Leu Asn Leu Pro Asn Phe Arg Asp Tyr Ala Val Asp Thr
    370                 375                 380
Ser Lys Glu Gly Ala Asn Val Lys Gln Asp Met Ile Val Trp Ser Asp
385                 390                 395                 400
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Tyr Leu Arg Asp Val Ile Lys Lys Asn Pro Asp Asn Phe Arg Leu Phe
405 410 415
Gly Pro Asp Glu Thr Met Ser Asn Arg Leu Tyr Gly Val Phe Glu Thr
420 425 430
Thr Asn Arg Gln Trp Met Glu Asp Ile His Pro Asp Ser Asp Gln Tyr
435 440 445
Glu Ala Pro Ala Gly Arg Val Leu Asp Ala Gln Leu Ser Glu His Gln
450 455 460
Ala Glu Gly Trp Leu Glu Gly Tyr Val Leu Thr Gly Arg His Gly Leu
465 470 475 480
Phe Ala Ser Tyr Glu Ala Phe Leu Arg Val Val Asp Ser Met Leu Thr
485 490 495
Gln His Phe Lys Trp Leu Arg Lys Ala Asn Glu Leu Asp Trp Arg Lys
500 505 510
Lys Tyr Pro Ser Leu Asn Ile Ile Ala Ala Ser Thr Val Phe Gln Gln
515 520 525
Asp His Asn Gly Tyr Thr His Gln Asp Pro Gly Ala Leu Thr His Leu
530 535 540
Ala Glu Lys Lys Pro Glu Tyr Ile Arg Glu Tyr Leu Pro Ala Asp Ala
545 550 555 560
Asn Thr Leu Leu Ala Val Gly Asp Val Ile Phe Arg Ser Gln Glu Lys
565 570 575
Ile Asn Tyr Val Val Thr Ser Lys His Pro Arg Gln Gln Trp Phe Ser
580 585 590
Ile Glu Glu Ala Lys Gln Leu Val Asp Asn Gly Leu Gly Ile Ile Asp
595 600 605
Trp Ala Ser Thr Asp Gln Gly Ser Glu Pro Asp Ile Val Phe Ala Ala
610 615 620
Ala Gly Thr Glu Pro Thr Leu Glu Thr Leu Ala Ala Ile Gln Leu Leu
625 630 635 640
His Asp Ser Phe Pro Glu Met Lys Ile Arg Phe Val Asn Val Val Asp
645 650 655
Ile Leu Lys Leu Arg Ser Pro Glu Lys Asp Pro Arg Gly Leu Ser Asp
660 665 670
Ala Glu Phe Asp His Tyr Phe Thr Lys Asp Lys Pro Val Val Phe Ala
675 680 685
Phe His Gly Tyr Glu Asp Leu Val Arg Asp Ile Phe Phe Asp Arg His
690 695 700
Asn His Asn Leu Tyr Val His Gly Tyr Arg Glu Asn Gly Asp Ile Thr
705 710 715 720
Thr Pro Phe Asp Val Arg Val Met Asn Gln Met Asp Arg Phe Asp Leu
725 730 735
Ala Lys Ser Ala Ile Ala Ala Gln Pro Ala Met Glu Asn Thr Gly Ala
740 745 750
Ala Phe Val Gln Ser Met Asp Asn Met Leu Ala Lys His Asn Ala Tyr

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                755                 760                 765
Ile Arg Asp Ala Gly Thr Asp Leu Pro Glu Val Asn Asp Trp Gln Trp
    770                 775                 780
Lys Gly Leu Lys
785
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<210> 56
<211> 2660
<212> DNA
<213> Bifidobacterium lactis

<220>
<221> CDS
<222> (128).. (2605)
<223>

<400> 56

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gcgccgaagc ccgtgcccaa cggaataaac aaatcgcaca tttatgtgca ggagtacagg    120
agcacac atg act aat cct gtt att ggt acc cca tgg cag aag ctg gat       169
       Met Thr Asn Pro Val Ile Gly Thr Pro Trp Gln Lys Leu Asp
       1               5                   10
cgt ccg gtt tcc gaa gag gcc atc gaa ggc atg gac aag tac tgg cgc      217
Arg Pro Val Ser Glu Glu Ala Ile Glu Gly Met Asp Lys Tyr Trp Arg
15                  20                  25                  30
gtc gcc aac tac atg tct atc ggc cag atc tac ctg cgt agc aac ccg      265
Val Ala Asn Tyr Met Ser Ile Gly Gln Ile Tyr Leu Arg Ser Asn Pro
                35                  40                  45
ctg atg aag gag ccc ttc acc cgc gat gac gtg aag cac cgt ctg gtc      313
Leu Met Lys Glu Pro Phe Thr Arg Asp Asp Val Lys His Arg Leu Val
            50                  55                  60
ggc cac tgg ggc acc acc ccg ggc ctg aac ttc ctt ctc gcc cac atc      361
Gly His Trp Gly Thr Thr Pro Gly Leu Asn Phe Leu Leu Ala His Ile
        65                  70                  75
aac cgc ctg atc gcc gat cac cag cag aac acc gtg ttc atc atg ggt      409
Asn Arg Leu Ile Ala Asp His Gln Gln Asn Thr Val Phe Ile Met Gly
    80                  85                  90
cct ggc cac ggc ggc cct gca ggt acc gct cag tcc tac atc gac ggc      457
Pro Gly His Gly Gly Pro Ala Gly Thr Ala Gln Ser Tyr Ile Asp Gly
95                  100                 105                 110
acc tac acc gag tac tac ccg aac atc acc aag gac gaa gct ggc ctg      505
Thr Tyr Thr Glu Tyr Tyr Pro Asn Ile Thr Lys Asp Glu Ala Gly Leu
                115                 120                 125
cag aag ttc ttc cgc cag ttc tcc tac ccg ggt ggc att cct tcc cac      553
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Gln Lys Phe Phe Arg Gln Phe Ser Tyr Pro Gly Gly Ile Pro Ser His
          130                 135                 140
ttc gct ccg gag acg ccg ggc tcc atc cac gaa ggc ggc gag ctg ggc   601
Phe Ala Pro Glu Thr Pro Gly Ser Ile His Glu Gly Gly Glu Leu Gly
        145                 150                 155
tac gcc ctg tcg cac gcc tac ggc gcg atc atg gac aac ccg agc ctc   649
Tyr Ala Leu Ser His Ala Tyr Gly Ala Ile Met Asp Asn Pro Ser Leu
    160                 165                 170
ttc gtc ccg tgc atc atc ggt gac ggc gaa gcc gag acc ggc cct ctg   697
Phe Val Pro Cys Ile Ile Gly Asp Gly Glu Ala Glu Thr Gly Pro Leu
175                 180                 185                 190
gcc acc ggc tgg cag tcc aac aag ctc gtc aac ccg cgc acc gac ggc   745
Ala Thr Gly Trp Gln Ser Asn Lys Leu Val Asn Pro Arg Thr Asp Gly
                195                 200                 205
atc gtc ctg ccg atc ctg cac ctc aac ggc tac aag atc gcc aac ccg   793
Ile Val Leu Pro Ile Leu His Leu Asn Gly Tyr Lys Ile Ala Asn Pro
            210                 215                 220
acg atc ctc gcc cgc atc tcc gac gag gag ctg cac gac ttc ttc cgc   841
Thr Ile Leu Ala Arg Ile Ser Asp Glu Glu Leu His Asp Phe Phe Arg
        225                 230                 235
ggc atg ggt tac cac ccg tac gag ttc gtc gcc ggc ttc gac aac gag   889
Gly Met Gly Tyr His Pro Tyr Glu Phe Val Ala Gly Phe Asp Asn Glu
    240                 245                 250
gat cac ctg tcg atc cac cgt cgc ttc gcc gag ctc ttc gag acc atc   937
Asp His Leu Ser Ile His Arg Arg Phe Ala Glu Leu Phe Glu Thr Ile
255                 260                 265                 270
ttc gac gag atc tgc gat atc aag gct gcg gct cag acc gac gac atg   985
Phe Asp Glu Ile Cys Asp Ile Lys Ala Ala Ala Gln Thr Asp Asp Met
                275                 280                 285
acc cgt ccg ttc tac ccg atg ctc atc ttc cgc acc ccg aag ggc tgg  1033
Thr Arg Pro Phe Tyr Pro Met Leu Ile Phe Arg Thr Pro Lys Gly Trp
            290                 295                 300
acc tgc ccg aag ttc atc gac ggc aag aag acc gaa ggc tcc tgg cgt  1081
Thr Cys Pro Lys Phe Ile Asp Gly Lys Lys Thr Glu Gly Ser Trp Arg
        305                 310                 315
gca cac cag gtc ccg ctg gct tcc gcc cgc gac acc gag gcc cac ttc  1129
Ala His Gln Val Pro Leu Ala Ser Ala Arg Asp Thr Glu Ala His Phe
    320                 325                 330
gaa gtc ctc aag ggc tgg atg gaa tcc tac aag ccg gag gag ctc ttc  1177
Glu Val Leu Lys Gly Trp Met Glu Ser Tyr Lys Pro Glu Glu Leu Phe
335                 340                 345                 350
aac gcc gac ggc tcc atc aag gag gac gtc acc gca ttc atg cct aag  1225
Asn Ala Asp Gly Ser Ile Lys Glu Asp Val Thr Ala Phe Met Pro Lys
                355                 360                 365
ggc gaa ctg cgc atc ggc gcc aac ccg aat gcc aac ggc ggc cgc atc  1273
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Gly Glu Leu Arg Ile Gly Ala Asn Pro Asn Ala Asn Gly Gly Arg Ile
            370                 375                 380
cgc gag gat ctg aag ctc cct gag ctc gat cag tac gag atc acc ggc    1321
Arg Glu Asp Leu Lys Leu Pro Glu Leu Asp Gln Tyr Glu Ile Thr Gly
        385                 390                 395
gtc aag gaa tac ggc cac ggt tgg ggc cag gtc gag gct ccg cgt tcc    1369
Val Lys Glu Tyr Gly His Gly Trp Gly Gln Val Glu Ala Pro Arg Ser
    400                 405                 410
ctc ggc gcg tac tgc cgc gac atc atc aag aac aac ccg gat tcg ttc    1417
Leu Gly Ala Tyr Cys Arg Asp Ile Ile Lys Asn Asn Pro Asp Ser Phe
415                 420                 425                 430
cgc gtc ttc gga cct gac gag acc gcg tcc aac cgt ctg aac gcg acc    1465
Arg Val Phe Gly Pro Asp Glu Thr Ala Ser Asn Arg Leu Asn Ala Thr
                435                 440                 445
tac gag gtc acc aag aag cag tgg gac aac gga tac ctc tcg gct ctc    1513
Tyr Glu Val Thr Lys Lys Gln Trp Asp Asn Gly Tyr Leu Ser Ala Leu
            450                 455                 460
gtc gac gag aac atg gcc gtc acc ggc cag gtt gtc gag cag ctc tcc    1561
Val Asp Glu Asn Met Ala Val Thr Gly Gln Val Val Glu Gln Leu Ser
        465                 470                 475
gag cat cag tgc gaa ggc ttc ctc gag gcc tac ctg ctc acc ggc cgt    1609
Glu His Gln Cys Glu Gly Phe Leu Glu Ala Tyr Leu Leu Thr Gly Arg
    480                 485                 490
cac ggc atc tgg agc tcc tac gag tcc ttc gtg cac gtg atc gac tcc    1657
His Gly Ile Trp Ser Ser Tyr Glu Ser Phe Val His Val Ile Asp Ser
495                 500                 505                 510
atg ctg aac cag cat gcg aag tgg ctc gag gcc acc gtc cgc gag atc    1705
Met Leu Asn Gln His Ala Lys Trp Leu Glu Ala Thr Val Arg Glu Ile
                515                 520                 525
ccg tgg cgt aag ccg atc tcc tcg gtg aac ctc ctg gtc tcc tcg cac    1753
Pro Trp Arg Lys Pro Ile Ser Ser Val Asn Leu Leu Val Ser Ser His
            530                 535                 540
gtg tgg cgt cag gat cac aac ggc ttc tcg cac cag gat ccg ggt gtg    1801
Val Trp Arg Gln Asp His Asn Gly Phe Ser His Gln Asp Pro Gly Val
        545                 550                 555
acc tcc gtc ctg ctg aac aag acg ttc aac aac gac cac gtg acg aac    1849
Thr Ser Val Leu Leu Asn Lys Thr Phe Asn Asn Asp His Val Thr Asn
    560                 565                 570
atc tac ttc gcg acc gat gcc aac atg ctg ctg gcc atc gcc gag aag    1897
Ile Tyr Phe Ala Thr Asp Ala Asn Met Leu Leu Ala Ile Ala Glu Lys
575                 580                 585                 590
tgc ttc aag tcc acc aac aag atc aac gca atc ttc gcc ggc aag cag    1945
Cys Phe Lys Ser Thr Asn Lys Ile Asn Ala Ile Phe Ala Gly Lys Gln
                595                 600                 605
ccg gcc gcg acg tgg atc acc ctc gac gag gta cgc gcc gag ctc gag    1993
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Pro Ala Ala Thr Trp Ile Thr Leu Asp Glu Val Arg Ala Glu Leu Glu
                610                 615                 620
gct ggt gcc gcc gag tgg aag tgg gct tcc aac gcc aag agc aac gac    2041
Ala Gly Ala Ala Glu Trp Lys Trp Ala Ser Asn Ala Lys Ser Asn Asp
        625                 630                 635
gag gtc cag gtt gtc ctc gcc gcc gcc ggc gac gtc ccg acc cag gag    2089
Glu Val Gln Val Val Leu Ala Ala Ala Gly Asp Val Pro Thr Gln Glu
    640                 645                 650
atc atg gcc gct tcc gat gcc ctc aac aag atg ggc atc aag ttc aag    2137
Ile Met Ala Ala Ser Asp Ala Leu Asn Lys Met Gly Ile Lys Phe Lys
655                 660                 665                 670
gtc gtc aac gtc gtg gac ctc atc aag ctg cag tcc tcg aag gag aac    2185
Val Val Asn Val Val Asp Leu Ile Lys Leu Gln Ser Ser Lys Glu Asn
                675                 680                 685
gac gag gcc atg tct gac gag gac ttc gcc gac ctg ttc acc gcg gac    2233
Asp Glu Ala Met Ser Asp Glu Asp Phe Ala Asp Leu Phe Thr Ala Asp
            690                 695                 700
aag ccg gtc ctc ttc gcc tac cac tcc tat gcc cag gac gtt cgt ggc    2281
Lys Pro Val Leu Phe Ala Tyr His Ser Tyr Ala Gln Asp Val Arg Gly
        705                 710                 715
ctc atc tac gac cgc ccg aac cac gac aac ttc acc gtt gtc gga tac    2329
Leu Ile Tyr Asp Arg Pro Asn His Asp Asn Phe Thr Val Val Gly Tyr
    720                 725                 730
aag gag cag ggc tcc acg acg acg ccg ttc gac atg gtg cgt gtc aac    2377
Lys Glu Gln Gly Ser Thr Thr Thr Pro Phe Asp Met Val Arg Val Asn
735                 740                 745                 750
gac atg gat cgc tac gcc ctt cag gcc aag gcc ctc gag ctc atc gac    2425
Asp Met Asp Arg Tyr Ala Leu Gln Ala Lys Ala Leu Glu Leu Ile Asp
                755                 760                 765
gcc gac aag tat gcc gac aag atc aac gag ctc aac gag ttc cgc aag    2473
Ala Asp Lys Tyr Ala Asp Lys Ile Asn Glu Leu Asn Glu Phe Arg Lys
            770                 775                 780
acc gcg ttc cag ttc gcc gtc gac aat ggc tat gac att cct gag ttc    2521
Thr Ala Phe Gln Phe Ala Val Asp Asn Gly Tyr Asp Ile Pro Glu Phe
        785                 790                 795
acc gat tgg gtg tac ccg gat gtc aag gtc gac gag acc tcc atg ctc    2569
Thr Asp Trp Val Tyr Pro Asp Val Lys Val Asp Glu Thr Ser Met Leu
    800                 805                 810
tcc gcc acc gcc gcg acc gcc ggc gac aac gag tga gcatagtctc         2615
Ser Ala Thr Ala Ala Thr Ala Gly Asp Asn Glu
815                 820                 825
atcgcttagc cgatgaaagg cccgggtgtc cgcacccggg ccttt                  2660
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<210> 57
<211> 825
<212> PRT
<213> Bifidobacterium lactic

<400> 57

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Met Thr Asn Pro Val Ile Gly Thr Pro Trp Gln Lys Leu Asp Arg Pro
1               5                  10                  15
Val Ser Glu Glu Ala Ile Glu Gly Met Asp Lys Tyr Trp Arg Val Ala
            20                  25                  30
Asn Tyr Met Ser Ile Gly Gln Ile Tyr Leu Arg Ser Asn Pro Leu Met
        35                  40                  45
Lys Glu Pro Phe Thr Arg Asp Asp Val Lys His Arg Leu Val Gly His
    50                  55                  60
Trp Gly Thr Thr Pro Gly Leu Asn Phe Leu Leu Ala His Ile Asn Arg
65                  70                  75                  80
Leu Ile Ala Asp His Gln Gln Asn Thr Val Phe Ile Met Gly Pro Gly
                85                  90                  95
His Gly Gly Pro Ala Gly Thr Ala Gln Ser Tyr Ile Asp Gly Thr Tyr
            100                 105                 110
Thr Glu Tyr Tyr Pro Asn Ile Thr Lys Asp Glu Ala Gly Leu Gln Lys
        115                 120                 125
Phe Phe Arg Gln Phe Ser Tyr Pro Gly Gly Ile Pro Ser His Phe Ala
    130                 135                 140
Pro Glu Thr Pro Gly Ser Ile His Glu Gly Gly Glu Leu Gly Tyr Ala
145                 150                 155                 160
Leu Ser His Ala Tyr Gly Ala Ile Met Asp Asn Pro Ser Leu Phe Val
                165                 170                 175
Pro Cys Ile Ile Gly Asp Gly Glu Ala Glu Thr Gly Pro Leu Ala Thr
            180                 185                 190
Gly Trp Gln Ser Asn Lys Leu Val Asn Pro Arg Thr Asp Gly Ile Val
        195                 200                 205
Leu Pro Ile Leu His Leu Asn Gly Tyr Lys Ile Ala Asn Pro Thr Ile
    210                 215                 220
Leu Ala Arg Ile Ser Asp Glu Glu Leu His Asp Phe Phe Arg Gly Met
225                 230                 235                 240
Gly Tyr His Pro Tyr Glu Phe Val Ala Gly Phe Asp Asn Glu Asp His
                245                 250                 255
Leu Ser Ile His Arg Arg Phe Ala Glu Leu Phe Glu Thr Ile Phe Asp
            260                 265                 270
Glu Ile Cys Asp Ile Lys Ala Ala Ala Gln Thr Asp Asp Met Thr Arg
        275                 280                 285
Pro Phe Tyr Pro Met Leu Ile Phe Arg Thr Pro Lys Gly Trp Thr Cys
    290                 295                 300
Pro Lys Phe Ile Asp Gly Lys Lys Thr Glu Gly Ser Trp Arg Ala His
```

305 310 315 320
Gln Val Pro Leu Ala Ser Ala Arg Asp Thr Glu Ala His Phe Glu Val
325 330 335
Leu Lys Gly Trp Met Glu Ser Tyr Lys Pro Glu Glu Leu Phe Asn Ala
340 345 350
Asp Gly Ser Ile Lys Glu Asp Val Thr Ala Phe Met Pro Lys Gly Glu
355 360 365
Leu Arg Ile Gly Ala Asn Pro Asn Ala Asn Gly Gly Arg Ile Arg Glu
370 375 380
Asp Leu Lys Leu Pro Glu Leu Asp Gln Tyr Glu Ile Thr Gly Val Lys
385 390 395 400
Glu Tyr Gly His Gly Trp Gly Gln Val Glu Ala Pro Arg Ser Leu Gly
405 410 415
Ala Tyr Cys Arg Asp Ile Ile Lys Asn Asn Pro Asp Ser Phe Arg Val
420 425 430
Phe Gly Pro Asp Glu Thr Ala Ser Asn Arg Leu Asn Ala Thr Tyr Glu
435 440 445
Val Thr Lys Lys Gln Trp Asp Asn Gly Tyr Leu Ser Ala Leu Val Asp
450 455 460
Glu Asn Met Ala Val Thr Gly Gln Val Val Glu Gln Leu Ser Glu His
465 470 475 480
Gln Cys Glu Gly Phe Leu Glu Ala Tyr Leu Leu Thr Gly Arg His Gly
485 490 495
Ile Trp Ser Ser Tyr Glu Ser Phe Val His Val Ile Asp Ser Met Leu
500 505 510
Asn Gln His Ala Lys Trp Leu Glu Ala Thr Val Arg Glu Ile Pro Trp
515 520 525
Arg Lys Pro Ile Ser Ser Val Asn Leu Leu Val Ser Ser His Val Trp
530 535 540
Arg Gln Asp His Asn Gly Phe Ser His Gln Asp Pro Gly Val Thr Ser
545 550 555 560
Val Leu Leu Asn Lys Thr Phe Asn Asn Asp His Val Thr Asn Ile Tyr
565 570 575
Phe Ala Thr Asp Ala Asn Met Leu Leu Ala Ile Ala Glu Lys Cys Phe
580 585 590
Lys Ser Thr Asn Lys Ile Asn Ala Ile Phe Ala Gly Lys Gln Pro Ala
595 600 605
Ala Thr Trp Ile Thr Leu Asp Glu Val Arg Ala Glu Leu Glu Ala Gly
610 615 620
Ala Ala Glu Trp Lys Trp Ala Ser Asn Ala Lys Ser Asn Asp Glu Val
625 630 635 640
Gln Val Val Leu Ala Ala Ala Gly Asp Val Pro Thr Gln Glu Ile Met
645 650 655
Ala Ala Ser Asp Ala Leu Asn Lys Met Gly Ile Lys Phe Lys Val Val
660 665 670

```
Asn Val Val Asp Leu Ile Lys Leu Gln Ser Ser Lys Glu Asn Asp Glu
        675                 680                 685
Ala Met Ser Asp Glu Asp Phe Ala Asp Leu Phe Thr Ala Asp Lys Pro
    690                 695                 700
Val Leu Phe Ala Tyr His Ser Tyr Ala Gln Asp Val Arg Gly Leu Ile
705                 710                 715                 720
Tyr Asp Arg Pro Asn His Asp Asn Phe Thr Val Val Gly Tyr Lys Glu
                725                 730                 735
Gln Gly Ser Thr Thr Thr Pro Phe Asp Met Val Arg Val Asn Asp Met
            740                 745                 750
Asp Arg Tyr Ala Leu Gln Ala Lys Ala Leu Glu Leu Ile Asp Ala Asp
        755                 760                 765
Lys Tyr Ala Asp Lys Ile Asn Glu Leu Asn Glu Phe Arg Lys Thr Ala
    770                 775                 780
Phe Gln Phe Ala Val Asp Asn Gly Tyr Asp Ile Pro Glu Phe Thr Asp
785                 790                 795                 800
Trp Val Tyr Pro Asp Val Lys Val Asp Glu Thr Ser Met Leu Ser Ala
                805                 810                 815
Thr Ala Ala Thr Ala Gly Asp Asn Glu
            820                 825
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<210> 58
<211> 23
<212> DNA
<213> Artificial sequence

<220>
<223> primer

<400> 58
ctgttaaggc agaaaccgtc gct 23

<210> 59
<211> 23
<212> DNA
<213> Artificial sequence

<220>
<223> primer

<400> 59
tcttggtgtc gaaagtgcac acc 23

<210> 60
<211> 20
<212> DNA
<213> Artificial sequence

<220>
<223> primer

<400> 60 20

cgcgaggtac cacctgtcac 20

<210> 61
<211> 20
<212> DNA
<213> Artificial sequence

<220>
<223> primer

<400> 61 20
caatccaggt accggcaacc 20

<210> 62
<211> 2760
<212> DNA
<213> Corynebacterium glutamicum

<220>
<221> CDS
<222> (1).. (2757)

<400> 62

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atg act gat ttt tta cgc gat gac atc agg ttc ctc ggt caa atc ctc      48
Met Thr Asp Phe Leu Arg Asp Asp Ile Arg Phe Leu Gly Gln Ile Leu
1               5                  10                  15
ggt gag gta att gcg gaa caa gaa ggc cag gag gtt tat gaa ctg gtc      96
Gly Glu Val Ile Ala Glu Gln Glu Gly Gln Glu Val Tyr Glu Leu Val
            20                  25                  30
gaa caa gcg cgc ctg act tct ttt gat atc gcc aag ggc aac gcc gaa     144
Glu Gln Ala Arg Leu Thr Ser Phe Asp Ile Ala Lys Gly Asn Ala Glu
        35                  40                  45
atg gat agc ctg gtt cag gtt ttc gac ggc att act cca gcc aag gca     192
```

```
Met Asp Ser Leu Val Gln Val Phe Asp Gly Ile Thr Pro Ala Lys Ala
    50                  55                  60
aca ccg att gct cgc gca ttt tcc cac ttc gct ctg ctg gct aac ctg      240
Thr Pro Ile Ala Arg Ala Phe Ser His Phe Ala Leu Leu Ala Asn Leu
65                  70                  75                  80
gcg gaa gac ctc tac gat gaa gag ctt cgt gaa cag gct ctc gat gca      288
Ala Glu Asp Leu Tyr Asp Glu Glu Leu Arg Glu Gln Ala Leu Asp Ala
                85                  90                  95
ggc gac acc cct ccg gac agc act ctt gat gcc acc tgg ctg aaa ctc      336
Gly Asp Thr Pro Pro Asp Ser Thr Leu Asp Ala Thr Trp Leu Lys Leu
            100                 105                 110
aat gag ggc aat gtt ggc gca gaa gct gtg gcc gat gtg ctg cgc aat      384
Asn Glu Gly Asn Val Gly Ala Glu Ala Val Ala Asp Val Leu Arg Asn
        115                 120                 125
gct gag gtg gcg ccg gtt ctg act gcg cac cca act gag act cgc cgc      432
Ala Glu Val Ala Pro Val Leu Thr Ala His Pro Thr Glu Thr Arg Arg
    130                 135                 140
cgc act gtt ttt gat gcg caa aag tgg atc acc acc cac atg cgt gaa      480
Arg Thr Val Phe Asp Ala Gln Lys Trp Ile Thr Thr His Met Arg Glu
145                 150                 155                 160
cgc cac gct ttg cag tct gcg gag cct acc gct cgt acg caa agc aag      528
Arg His Ala Leu Gln Ser Ala Glu Pro Thr Ala Arg Thr Gln Ser Lys
                165                 170                 175
ttg gat gag atc gag aag aac atc cgc cgt cgc atc acc att ttg tgg      576
Leu Asp Glu Ile Glu Lys Asn Ile Arg Arg Arg Ile Thr Ile Leu Trp
            180                 185                 190
cag acc gcg ttg att cgt gtg gcc cgc cca cgt atc gag gac gag atc      624
Gln Thr Ala Leu Ile Arg Val Ala Arg Pro Arg Ile Glu Asp Glu Ile
        195                 200                 205
gaa gta ggg ctg cgc tac tac aag ctg agc ctt ttg gaa gag att cca      672
Glu Val Gly Leu Arg Tyr Tyr Lys Leu Ser Leu Leu Glu Glu Ile Pro
    210                 215                 220
cgt atc aac cgt gat gtg gct gtt gag ctt cgt gag cgt ttc ggc gag      720
Arg Ile Asn Arg Asp Val Ala Val Glu Leu Arg Glu Arg Phe Gly Glu
225                 230                 235                 240
ggt gtt cct ttg aag ccc gtg gtc aag cca ggt tcc tgg att ggt gga      768
Gly Val Pro Leu Lys Pro Val Val Lys Pro Gly Ser Trp Ile Gly Gly
                245                 250                 255
gac cac gac ggt aac cct tat gtc acc gcg gaa aca gtt gag tat tcc      816
Asp His Asp Gly Asn Pro Tyr Val Thr Ala Glu Thr Val Glu Tyr Ser
            260                 265                 270
act cac cgc gct gcg gaa acc gtg ctc aag tac tat gca cgc cag ctg      864
Thr His Arg Ala Ala Glu Thr Val Leu Lys Tyr Tyr Ala Arg Gln Leu
        275                 280                 285
cat tcc ctc gag cat gag ctc agc ctg tcg gac cgc atg aat aag gtc      912
```

```
His Ser Leu Glu His Glu Leu Ser Leu Ser Asp Arg Met Asn Lys Val
290                 295                 300
acc ccg cag ctg ctt gcg ctg gca gat gca ggg cac aac gac gtg cca   960
Thr Pro Gln Leu Leu Ala Leu Ala Asp Ala Gly His Asn Asp Val Pro
305                 310                 315                 320
agc cgc gtg gat gag cct tat cga cgc gcc gtc cat ggc gtt cgc gga   1008
Ser Arg Val Asp Glu Pro Tyr Arg Arg Ala Val His Gly Val Arg Gly
                325                 330                 335
cgt atc ctc gcg acg acg gcc gag ctg atc ggc gag gac gcc gtt gag   1056
Arg Ile Leu Ala Thr Thr Ala Glu Leu Ile Gly Glu Asp Ala Val Glu
            340                 345                 350
ggc gtg tgg ttc aag gtc ttt act cca tac gca tct ccg gaa gaa ttc   1104
Gly Val Trp Phe Lys Val Phe Thr Pro Tyr Ala Ser Pro Glu Glu Phe
        355                 360                 365
tta aac gat gcg ttg acc att gat cat tct ctg cgt gaa tcc aag gac   1152
Leu Asn Asp Ala Leu Thr Ile Asp His Ser Leu Arg Glu Ser Lys Asp
    370                 375                 380
gtt ctc att gcc gat gat cgt ttg tct gtg ctg att tct gcc atc gag   1200
Val Leu Ile Ala Asp Asp Arg Leu Ser Val Leu Ile Ser Ala Ile Glu
385                 390                 395                 400
agc ttt gga ttc aac ctt tac gca ctg gat ctg cgc caa aac tcc gaa   1248
Ser Phe Gly Phe Asn Leu Tyr Ala Leu Asp Leu Arg Gln Asn Ser Glu
                405                 410                 415
agc tac gag gac gtc ctc acc gag ctt ttc gaa cgc gcc caa gtc acc   1296
Ser Tyr Glu Asp Val Leu Thr Glu Leu Phe Glu Arg Ala Gln Val Thr
            420                 425                 430
gca aac tac cgc gag ctg tct gaa gca gag aag ctt gag gtg ctg ctg   1344
Ala Asn Tyr Arg Glu Leu Ser Glu Ala Glu Lys Leu Glu Val Leu Leu
        435                 440                 445
aag gaa ctg cgc agc cct cgt ccg ctg atc ccg cac ggt tca gat gaa   1392
Lys Glu Leu Arg Ser Pro Arg Pro Leu Ile Pro His Gly Ser Asp Glu
    450                 455                 460
tac agc gag gtc acc gac cgc gag ctc ggc atc ttc cgc acc gcg tcg   1440
Tyr Ser Glu Val Thr Asp Arg Glu Leu Gly Ile Phe Arg Thr Ala Ser
465                 470                 475                 480
gag gct gtt aag aaa ttc ggg cca cgg atg gtg cct cac tgc atc atc   1488
Glu Ala Val Lys Lys Phe Gly Pro Arg Met Val Pro His Cys Ile Ile
                485                 490                 495
tcc atg gca tca tcg gtc acc gat gtg ctc gag ccg atg gtg ttg ctc   1536
Ser Met Ala Ser Ser Val Thr Asp Val Leu Glu Pro Met Val Leu Leu
            500                 505                 510
aag gaa ttc gga ctc atc gca gcc aac ggc gac aac cca cgc ggc acc   1584
Lys Glu Phe Gly Leu Ile Ala Ala Asn Gly Asp Asn Pro Arg Gly Thr
        515                 520                 525
gtc gat gtc atc cca ctg ttc gaa acc atc gaa gat ctc cag gcc ggc   1632
```

```
Val Asp Val Ile Pro Leu Phe Glu Thr Ile Glu Asp Leu Gln Ala Gly
    530                 535                 540
gcc gga atc ctc gac gaa ctg tgg aaa att gat ctc tac cgc aac tac     1680
Ala Gly Ile Leu Asp Glu Leu Trp Lys Ile Asp Leu Tyr Arg Asn Tyr
545                 550                 555                 560
ctc ctg cag cgc gac aac gtc cag gaa gtc atg ctc ggt tac tcc gat     1728
Leu Leu Gln Arg Asp Asn Val Gln Glu Val Met Leu Gly Tyr Ser Asp
                565                 570                 575
tcc aac aag gat ggc gga tat ttc tcc gca aac tgg gcg ctt tac gac     1776
Ser Asn Lys Asp Gly Gly Tyr Phe Ser Ala Asn Trp Ala Leu Tyr Asp
            580                 585                 590
gcg gaa ctg cag ctc gtc gaa cta tgc cga tca gcc ggg gtc aag ctt     1824
Ala Glu Leu Gln Leu Val Glu Leu Cys Arg Ser Ala Gly Val Lys Leu
        595                 600                 605
cgc ctg ttc cac ggc cgt ggt ggc acc gtc ggc cgc ggt ggc gga cct     1872
Arg Leu Phe His Gly Arg Gly Gly Thr Val Gly Arg Gly Gly Gly Pro
    610                 615                 620
tcc tac gac gcg att ctt gcc cag ccc agg ggg gct gtc caa ggt tcc     1920
Ser Tyr Asp Ala Ile Leu Ala Gln Pro Arg Gly Ala Val Gln Gly Ser
625                 630                 635                 640
gtg cgc atc acc gag cag ggc gag atc atc tcc gct aag tac ggc aac     1968
Val Arg Ile Thr Glu Gln Gly Glu Ile Ile Ser Ala Lys Tyr Gly Asn
                645                 650                 655
ccc gaa acc gcg cgc cga aac ctc gaa gcc ctg gtc tca gcc acg ctt     2016
Pro Glu Thr Ala Arg Arg Asn Leu Glu Ala Leu Val Ser Ala Thr Leu
            660                 665                 670
gag gca tcg ctt ctc gac gtc tcc gaa ctc acc gat cac caa cgc gcg     2064
Glu Ala Ser Leu Leu Asp Val Ser Glu Leu Thr Asp His Gln Arg Ala
        675                 680                 685
tac gac atc atg agt gag atc tct gag ctc agc ttg aag aag tac gcc     2112
Tyr Asp Ile Met Ser Glu Ile Ser Glu Leu Ser Leu Lys Lys Tyr Ala
    690                 695                 700
tcc ttg gtg cac gag gat caa ggc ttc atc gat tac ttc acc cag tcc     2160
Ser Leu Val His Glu Asp Gln Gly Phe Ile Asp Tyr Phe Thr Gln Ser
705                 710                 715                 720
acg ccg ctg cag gag att gga tcc ctc aac atc gga tcc agg cct tcc     2208
Thr Pro Leu Gln Glu Ile Gly Ser Leu Asn Ile Gly Ser Arg Pro Ser
                725                 730                 735
tca cgc aag cag acc tcc tcg gtg gaa gat ttg cga gcc atc cca tgg     2256
Ser Arg Lys Gln Thr Ser Ser Val Glu Asp Leu Arg Ala Ile Pro Trp
            740                 745                 750
gtg ctc agc tgg tca cag tct cgt gtc atg ctg cca ggc tgg ttt ggt     2304
Val Leu Ser Trp Ser Gln Ser Arg Val Met Leu Pro Gly Trp Phe Gly
        755                 760                 765
gtc gga acc gca tta gag cag tgg att ggc gaa ggg gag cag gcc acc     2352
```

```
Val Gly Thr Ala Leu Glu Gln Trp Ile Gly Glu Gly Glu Gln Ala Thr
    770                 775                 780
caa cgc att gcc gag ctg caa aca ctc aat gag tcc tgg cca ttt ttc     2400
Gln Arg Ile Ala Glu Leu Gln Thr Leu Asn Glu Ser Trp Pro Phe Phe
785                 790                 795                 800
acc tca gtg ttg gat aac atg gct cag gtg atg tcc aag gca gag ctg     2448
Thr Ser Val Leu Asp Asn Met Ala Gln Val Met Ser Lys Ala Glu Leu
                805                 810                 815
cgt ttg gca aag ctc tac gca gac ctg atc cca gat acg gaa gta gcc     2496
Arg Leu Ala Lys Leu Tyr Ala Asp Leu Ile Pro Asp Thr Glu Val Ala
            820                 825                 830
gag cga gtc tat tcc gtc atc cgc gag gag tac ttc ctg acc aag aag     2544
Glu Arg Val Tyr Ser Val Ile Arg Glu Glu Tyr Phe Leu Thr Lys Lys
        835                 840                 845
atg ttc tgc gta atc acc ggc tct gat gat ctg ctt gat gac aac cca     2592
Met Phe Cys Val Ile Thr Gly Ser Asp Asp Leu Leu Asp Asp Asn Pro
    850                 855                 860
ctt ctc gca cgc tct gtc cag cgc cga tac ccc tac ctg ctt cca ctc     2640
Leu Leu Ala Arg Ser Val Gln Arg Arg Tyr Pro Tyr Leu Leu Pro Leu
865                 870                 875                 880
aac gtg atc cag gta gag atg atg cga cgc tac cga aaa ggc gac caa     2688
Asn Val Ile Gln Val Glu Met Met Arg Arg Tyr Arg Lys Gly Asp Gln
                885                 890                 895
agc gag caa gtg tcc cgc aac att cag ctg acc atg aac ggt ctt tcc     2736
Ser Glu Gln Val Ser Arg Asn Ile Gln Leu Thr Met Asn Gly Leu Ser
            900                 905                 910
act gcg ctg cgc aac tcc ggc tag                                    2760
Thr Ala Leu Arg Asn Ser Gly
        915
```

<210> 63
<211> 919
<212> PRT
<213> Corynebacterium glutamicum

<400> 63

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Met Thr Asp Phe Leu Arg Asp Asp Ile Arg Phe Leu Gly Gln Ile Leu
1               5                   10                  15
Gly Glu Val Ile Ala Glu Gln Glu Gly Gln Glu Val Tyr Glu Leu Val
            20                  25                  30
Glu Gln Ala Arg Leu Thr Ser Phe Asp Ile Ala Lys Gly Asn Ala Glu
        35                  40                  45
Met Asp Ser Leu Val Gln Val Phe Asp Gly Ile Thr Pro Ala Lys Ala
    50                  55                  60
```

Thr Pro Ile Ala Arg Ala Phe Ser His Phe Ala Leu Leu Ala Asn Leu
65 70 75 80
Ala Glu Asp Leu Tyr Asp Glu Glu Leu Arg Glu Gln Ala Leu Asp Ala
85 90 95
Gly Asp Thr Pro Pro Asp Ser Thr Leu Asp Ala Thr Trp Leu Lys Leu
100 105 110
Asn Glu Gly Asn Val Gly Ala Glu Ala Val Ala Asp Val Leu Arg Asn
115 120 125
Ala Glu Val Ala Pro Val Leu Thr Ala His Pro Thr Glu Thr Arg Arg
130 135 140
Arg Thr Val Phe Asp Ala Gln Lys Trp Ile Thr Thr His Met Arg Glu
145 150 155 160
Arg His Ala Leu Gln Ser Ala Glu Pro Thr Ala Arg Thr Gln Ser Lys
165 170 175
Leu Asp Glu Ile Glu Lys Asn Ile Arg Arg Arg Ile Thr Ile Leu Trp
180 185 190
Gln Thr Ala Leu Ile Arg Val Ala Arg Pro Arg Ile Glu Asp Glu Ile
195 200 205
Glu Val Gly Leu Arg Tyr Tyr Lys Leu Ser Leu Leu Glu Glu Ile Pro
210 215 220
Arg Ile Asn Arg Asp Val Ala Val Glu Leu Arg Glu Arg Phe Gly Glu
225 230 235 240
Gly Val Pro Leu Lys Pro Val Val Lys Pro Gly Ser Trp Ile Gly Gly
245 250 255
Asp His Asp Gly Asn Pro Tyr Val Thr Ala Glu Thr Val Glu Tyr Ser
260 265 270
Thr His Arg Ala Ala Glu Thr Val Leu Lys Tyr Tyr Ala Arg Gln Leu
275 280 285
His Ser Leu Glu His Glu Leu Ser Leu Ser Asp Arg Met Asn Lys Val
290 295 300
Thr Pro Gln Leu Leu Ala Leu Ala Asp Ala Gly His Asn Asp Val Pro
305 310 315 320
Ser Arg Val Asp Glu Pro Tyr Arg Arg Ala Val His Gly Val Arg Gly
325 330 335
Arg Ile Leu Ala Thr Thr Ala Glu Leu Ile Gly Glu Asp Ala Val Glu
340 345 350
Gly Val Trp Phe Lys Val Phe Thr Pro Tyr Ala Ser Pro Glu Glu Phe
355 360 365
Leu Asn Asp Ala Leu Thr Ile Asp His Ser Leu Arg Glu Ser Lys Asp
370 375 380
Val Leu Ile Ala Asp Asp Arg Leu Ser Val Leu Ile Ser Ala Ile Glu
385 390 395 400
Ser Phe Gly Phe Asn Leu Tyr Ala Leu Asp Leu Arg Gln Asn Ser Glu
405 410 415
Ser Tyr Glu Asp Val Leu Thr Glu Leu Phe Glu Arg Ala Gln Val Thr

420 425 430

Ala Asn Tyr Arg Glu Leu Ser Glu Ala Glu Lys Leu Glu Val Leu Leu
435 440 445

Lys Glu Leu Arg Ser Pro Arg Pro Leu Ile Pro His Gly Ser Asp Glu
450 455 460

Tyr Ser Glu Val Thr Asp Arg Glu Leu Gly Ile Phe Arg Thr Ala Ser
465 470 475 480

Glu Ala Val Lys Lys Phe Gly Pro Arg Met Val Pro His Cys Ile Ile
485 490 495

Ser Met Ala Ser Ser Val Thr Asp Val Leu Glu Pro Met Val Leu Leu
500 505 510

Lys Glu Phe Gly Leu Ile Ala Ala Asn Gly Asp Asn Pro Arg Gly Thr
515 520 525

Val Asp Val Ile Pro Leu Phe Glu Thr Ile Glu Asp Leu Gln Ala Gly
530 535 540

Ala Gly Ile Leu Asp Glu Leu Trp Lys Ile Asp Leu Tyr Arg Asn Tyr
545 550 555 560

Leu Leu Gln Arg Asp Asn Val Gln Glu Val Met Leu Gly Tyr Ser Asp
565 570 575

Ser Asn Lys Asp Gly Gly Tyr Phe Ser Ala Asn Trp Ala Leu Tyr Asp
580 585 590

Ala Glu Leu Gln Leu Val Glu Leu Cys Arg Ser Ala Gly Val Lys Leu
595 600 605

Arg Leu Phe His Gly Arg Gly Gly Thr Val Gly Arg Gly Gly Gly Pro
610 615 620

Ser Tyr Asp Ala Ile Leu Ala Gln Pro Arg Gly Ala Val Gln Gly Ser
625 630 635 640

Val Arg Ile Thr Glu Gln Gly Glu Ile Ile Ser Ala Lys Tyr Gly Asn
645 650 655

Pro Glu Thr Ala Arg Arg Asn Leu Glu Ala Leu Val Ser Ala Thr Leu
660 665 670

Glu Ala Ser Leu Leu Asp Val Ser Glu Leu Thr Asp His Gln Arg Ala
675 680 685

Tyr Asp Ile Met Ser Glu Ile Ser Glu Leu Ser Leu Lys Lys Tyr Ala
690 695 700

Ser Leu Val His Glu Asp Gln Gly Phe Ile Asp Tyr Phe Thr Gln Ser
705 710 715 720

Thr Pro Leu Gln Glu Ile Gly Ser Leu Asn Ile Gly Ser Arg Pro Ser
725 730 735

Ser Arg Lys Gln Thr Ser Ser Val Glu Asp Leu Arg Ala Ile Pro Trp
740 745 750

Val Leu Ser Trp Ser Gln Ser Arg Val Met Leu Pro Gly Trp Phe Gly
755 760 765

Val Gly Thr Ala Leu Glu Gln Trp Ile Gly Glu Gly Glu Gln Ala Thr
770 775 780

```
Gln Arg Ile Ala Glu Leu Gln Thr Leu Asn Glu Ser Trp Pro Phe Phe
785                 790                 795                 800
Thr Ser Val Leu Asp Asn Met Ala Gln Val Met Ser Lys Ala Glu Leu
                805                 810                 815
Arg Leu Ala Lys Leu Tyr Ala Asp Leu Ile Pro Asp Thr Glu Val Ala
            820                 825                 830
Glu Arg Val Tyr Ser Val Ile Arg Glu Glu Tyr Phe Leu Thr Lys Lys
        835                 840                 845
Met Phe Cys Val Ile Thr Gly Ser Asp Asp Leu Leu Asp Asp Asn Pro
    850                 855                 860
Leu Leu Ala Arg Ser Val Gln Arg Arg Tyr Pro Tyr Leu Leu Pro Leu
865                 870                 875                 880
Asn Val Ile Gln Val Glu Met Met Arg Arg Tyr Arg Lys Gly Asp Gln
                885                 890                 895
Ser Glu Gln Val Ser Arg Asn Ile Gln Leu Thr Met Asn Gly Leu Ser
            900                 905                 910
Thr Ala Leu Arg Asn Ser Gly
        915
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<210> 64
<211> 3423
<212> DNA
<213> Corynebacterium glutamicum

<220>
<221> CDS
<222> (1).. (3420)

<400> 64

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gtg tcg act cac aca tct tca acg ctt cca gca ttc aaa aag atc ttg      48
Val Ser Thr His Thr Ser Ser Thr Leu Pro Ala Phe Lys Lys Ile Leu
1               5                   10                  15
gta gca aac cgc ggc gaa atc gcg gtc cgt gct ttc cgt gca gca ctc      96
Val Ala Asn Arg Gly Glu Ile Ala Val Arg Ala Phe Arg Ala Ala Leu
            20                  25                  30
gaa acc ggt gca gcc acg gta gct att tac ccc cgt gaa gat cgg gga     144
Glu Thr Gly Ala Ala Thr Val Ala Ile Tyr Pro Arg Glu Asp Arg Gly
        35                  40                  45
tca ttc cac cgc tct ttt gct tct gaa gct gtc cgc att ggt acc gaa     192
Ser Phe His Arg Ser Phe Ala Ser Glu Ala Val Arg Ile Gly Thr Glu
    50                  55                  60
ggc tca cca gtc aag gcg tac ctg gac atc gat gaa att atc ggt gca     240
Gly Ser Pro Val Lys Ala Tyr Leu Asp Ile Asp Glu Ile Ile Gly Ala
65                  70                  75                  80
```

```
gct aaa aaa gtt aaa gca gat gcc att tac ccg gga tac ggc ttc ctg      288
Ala Lys Lys Val Lys Ala Asp Ala Ile Tyr Pro Gly Tyr Gly Phe Leu
                85                  90                  95

tct gaa aat gcc cag ctt gcc cgc gag tgt gcg gaa aac ggc att act      336
Ser Glu Asn Ala Gln Leu Ala Arg Glu Cys Ala Glu Asn Gly Ile Thr
            100                 105                 110

ttt att ggc cca acc cca gag gtt ctt gat ctc acc ggt gat aag tct      384
Phe Ile Gly Pro Thr Pro Glu Val Leu Asp Leu Thr Gly Asp Lys Ser
        115                 120                 125

cgc gcg gta acc gcc gcg aag aag gct ggt ctg cca gtt ttg gcg gaa      432
Arg Ala Val Thr Ala Ala Lys Lys Ala Gly Leu Pro Val Leu Ala Glu
    130                 135                 140

tcc acc ccg agc aaa aac atc gat gag atc gtt aaa agc gct gaa ggc      480
Ser Thr Pro Ser Lys Asn Ile Asp Glu Ile Val Lys Ser Ala Glu Gly
145                 150                 155                 160

cag act tac ccc atc ttt gtg aag gca gtt gcc ggt ggt ggc gga cgc      528
Gln Thr Tyr Pro Ile Phe Val Lys Ala Val Ala Gly Gly Gly Gly Arg
                165                 170                 175

ggt atg cgt ttt gtt gct tca cct gat gag ctt cgc aaa tta gca aca      576
Gly Met Arg Phe Val Ala Ser Pro Asp Glu Leu Arg Lys Leu Ala Thr
            180                 185                 190

gaa gca tct cgt gaa gct gaa gcg gct ttc ggc gat ggc gcg gta tat      624
Glu Ala Ser Arg Glu Ala Glu Ala Ala Phe Gly Asp Gly Ala Val Tyr
        195                 200                 205

gtc gaa cgt gct gtg att aac cct cag cat att gaa gtg cag atc ctt      672
Val Glu Arg Ala Val Ile Asn Pro Gln His Ile Glu Val Gln Ile Leu
    210                 215                 220

ggc gat cac act gga gaa gtt gta cac ctt tat gaa cgt gac tgc tca      720
Gly Asp His Thr Gly Glu Val Val His Leu Tyr Glu Arg Asp Cys Ser
225                 230                 235                 240

ctg cag cgt cgt cac caa aaa gtt gtc gaa att gcg cca gca cag cat      768
Leu Gln Arg Arg His Gln Lys Val Val Glu Ile Ala Pro Ala Gln His
                245                 250                 255

ttg gat cca gaa ctg cgt gat cgc att tgt gcg gat gca gta aag ttc      816
Leu Asp Pro Glu Leu Arg Asp Arg Ile Cys Ala Asp Ala Val Lys Phe
            260                 265                 270

tgc cgc tcc att ggt tac cag ggc gcg gga acc gtg gaa ttc ttg gtc      864
Cys Arg Ser Ile Gly Tyr Gln Gly Ala Gly Thr Val Glu Phe Leu Val
        275                 280                 285

gat gaa aag ggc aac cac gtc ttc atc gaa atg aac cca cgt atc cag      912
Asp Glu Lys Gly Asn His Val Phe Ile Glu Met Asn Pro Arg Ile Gln
    290                 295                 300

gtt gag cac acc gtg act gaa gaa gtc acc gag gtg gac ctg gtg aag      960
Val Glu His Thr Val Thr Glu Glu Val Thr Glu Val Asp Leu Val Lys
305                 310                 315                 320
```

```
gcg cag atg cgc ttg gct gct ggt gca acc ttg aag gaa ttg ggt ctg     1008
Ala Gln Met Arg Leu Ala Ala Gly Ala Thr Leu Lys Glu Leu Gly Leu
                325                 330                 335
acc caa gat aag atc aag acc cac ggt gca gca ctg cag tgc cgc atc     1056
Thr Gln Asp Lys Ile Lys Thr His Gly Ala Ala Leu Gln Cys Arg Ile
            340                 345                 350
acc acg gaa gat cca aac aac ggc ttc cgc cca gat acc gga act atc     1104
Thr Thr Glu Asp Pro Asn Asn Gly Phe Arg Pro Asp Thr Gly Thr Ile
        355                 360                 365
acc gcg tac cgc tca cca ggc gga gct ggc gtt cgt ctt gac ggt gca     1152
Thr Ala Tyr Arg Ser Pro Gly Gly Ala Gly Val Arg Leu Asp Gly Ala
    370                 375                 380
gct cag ctc ggt ggc gaa atc acc gca cac ttt gac tcc atg ctg gtg     1200
Ala Gln Leu Gly Gly Glu Ile Thr Ala His Phe Asp Ser Met Leu Val
385                 390                 395                 400
aaa atg acc tgc cgt ggt tcc gac ttt gaa act gct gtt gct cgt gca     1248
Lys Met Thr Cys Arg Gly Ser Asp Phe Glu Thr Ala Val Ala Arg Ala
                405                 410                 415
cag cgc gcg ttg gct gag ttc acc gtg tct ggt gtt gca acc aac att     1296
Gln Arg Ala Leu Ala Glu Phe Thr Val Ser Gly Val Ala Thr Asn Ile
            420                 425                 430
ggt ttc ttg cgt gcg ttg ctg cgg gaa gag gac ttc act tcc aag cgc     1344
Gly Phe Leu Arg Ala Leu Leu Arg Glu Glu Asp Phe Thr Ser Lys Arg
        435                 440                 445
atc gcc acc gga ttc att gcc gat cac ccg cac ctc ctt cag gct cca     1392
Ile Ala Thr Gly Phe Ile Ala Asp His Pro His Leu Leu Gln Ala Pro
    450                 455                 460
cct gct gat gat gag cag gga cgc atc ctg gat tac ttg gca gat gtc     1440
Pro Ala Asp Asp Glu Gln Gly Arg Ile Leu Asp Tyr Leu Ala Asp Val
465                 470                 475                 480
acc gtg aac aag cct cat ggt gtg cgt cca aag gat gtt gca gct cct     1488
Thr Val Asn Lys Pro His Gly Val Arg Pro Lys Asp Val Ala Ala Pro
                485                 490                 495
atc gat aag ctg cct aac atc aag gat ctg cca ctg cca cgc ggt tcc     1536
Ile Asp Lys Leu Pro Asn Ile Lys Asp Leu Pro Leu Pro Arg Gly Ser
            500                 505                 510
cgt gac cgc ctg aag cag ctt ggc cca gcc gcg ttt gct cgt gat ctc     1584
Arg Asp Arg Leu Lys Gln Leu Gly Pro Ala Ala Phe Ala Arg Asp Leu
        515                 520                 525
cgt gag cag gac gca ctg gca gtt act gat acc acc ttc cgc gat gca     1632
Arg Glu Gln Asp Ala Leu Ala Val Thr Asp Thr Thr Phe Arg Asp Ala
    530                 535                 540
cac cag tct ttg ctt gcg acc cga gtc cgc tca ttc gca ctg aag cct     1680
His Gln Ser Leu Leu Ala Thr Arg Val Arg Ser Phe Ala Leu Lys Pro
545                 550                 555                 560
```

```
gcg gca gag gcc gtc gca aag ctg act cct gag ctt ttg tcc gtg gag     1728
Ala Ala Glu Ala Val Ala Lys Leu Thr Pro Glu Leu Leu Ser Val Glu
                565                 570                 575

gcc tgg ggc ggc gcg acc tac gat gtg gcg atg cgt ttc ctc ttt gag     1776
Ala Trp Gly Gly Ala Thr Tyr Asp Val Ala Met Arg Phe Leu Phe Glu
            580                 585                 590

gat ccg tgg gac agg ctc gac gag ctg cgc gag gcg atg ccg aat gta     1824
Asp Pro Trp Asp Arg Leu Asp Glu Leu Arg Glu Ala Met Pro Asn Val
        595                 600                 605

aac att cag atg ctg ctt cgc ggc cgc aac acc gtg gga tac acc ccg     1872
Asn Ile Gln Met Leu Leu Arg Gly Arg Asn Thr Val Gly Tyr Thr Pro
    610                 615                 620

tac cca gac tcc gtc tgc cgc gcg ttt gtt aag gaa gct gcc agc tcc     1920
Tyr Pro Asp Ser Val Cys Arg Ala Phe Val Lys Glu Ala Ala Ser Ser
625                 630                 635                 640

ggc gtg gac atc ttc cgc atc ttc gac gcg ctt aac gac gtc tcc cag     1968
Gly Val Asp Ile Phe Arg Ile Phe Asp Ala Leu Asn Asp Val Ser Gln
                645                 650                 655

atg cgt cca gca atc gac gca gtc ctg gag acc aac acc gcg gta gcc     2016
Met Arg Pro Ala Ile Asp Ala Val Leu Glu Thr Asn Thr Ala Val Ala
            660                 665                 670

gag gtg gct atg gct tat tct ggt gat ctc tct gat cca aat gaa aag     2064
Glu Val Ala Met Ala Tyr Ser Gly Asp Leu Ser Asp Pro Asn Glu Lys
        675                 680                 685

ctc tac acc ctg gat tac tac cta aag atg gca gag gag atc gtc aag     2112
Leu Tyr Thr Leu Asp Tyr Tyr Leu Lys Met Ala Glu Glu Ile Val Lys
    690                 695                 700

tct ggc gct cac atc ttg gcc att aag gat atg gct ggt ctg ctt cgc     2160
Ser Gly Ala His Ile Leu Ala Ile Lys Asp Met Ala Gly Leu Leu Arg
705                 710                 715                 720

cca gct gcg gta acc aag ctg gtc acc gca ctg cgc cgt gaa ttc gat     2208
Pro Ala Ala Val Thr Lys Leu Val Thr Ala Leu Arg Arg Glu Phe Asp
                725                 730                 735

ctg cca gtg cac gtg cac acc cac gac act gcg ggt ggc cag ctg gca     2256
Leu Pro Val His Val His Thr His Asp Thr Ala Gly Gly Gln Leu Ala
            740                 745                 750

acc tac ttt gct gca gct caa gct ggt gca gat gct gtt gac ggt gct     2304
Thr Tyr Phe Ala Ala Ala Gln Ala Gly Ala Asp Ala Val Asp Gly Ala
        755                 760                 765

tcc gca cca ctg tct ggc acc acc tcc cag cca tcc ctg tct gcc att     2352
Ser Ala Pro Leu Ser Gly Thr Thr Ser Gln Pro Ser Leu Ser Ala Ile
    770                 775                 780

gtt gct gca ttc gcg cac acc cgt cgc gat acc ggt ttg agc ctc gag     2400
Val Ala Ala Phe Ala His Thr Arg Arg Asp Thr Gly Leu Ser Leu Glu
785                 790                 795                 800
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gct gtt tct gac ctc gag ccg tac tgg gaa gca gtg cgc gga ctg tac      2448
Ala Val Ser Asp Leu Glu Pro Tyr Trp Glu Ala Val Arg Gly Leu Tyr
                805                 810                 815

ctg cca ttt gag tct gga acc cca ggc cca acc ggt cgc gtc tac cgc      2496
Leu Pro Phe Glu Ser Gly Thr Pro Gly Pro Thr Gly Arg Val Tyr Arg
            820                 825                 830

cac gaa atc cca ggc gga cag ttg tcc aac ctg cgt gca cag gcc acc      2544
His Glu Ile Pro Gly Gly Gln Leu Ser Asn Leu Arg Ala Gln Ala Thr
        835                 840                 845

gca ctg ggc ctt gcg gat cgt ttc gaa ctc atc gaa gac aac tac gca      2592
Ala Leu Gly Leu Ala Asp Arg Phe Glu Leu Ile Glu Asp Asn Tyr Ala
    850                 855                 860

gcc gtt aat gag atg ctg gga cgc cca acc aag gtc acc cca tcc tcc      2640
Ala Val Asn Glu Met Leu Gly Arg Pro Thr Lys Val Thr Pro Ser Ser
865                 870                 875                 880

aag gtt gtt ggc gac ctc gca ctc cac ctc gtt ggt gcg ggt gtg gat      2688
Lys Val Val Gly Asp Leu Ala Leu His Leu Val Gly Ala Gly Val Asp
                885                 890                 895

cca gca gac ttt gct gcc gat cca caa aag tac gac atc cca gac tct      2736
Pro Ala Asp Phe Ala Ala Asp Pro Gln Lys Tyr Asp Ile Pro Asp Ser
            900                 905                 910

gtc atc gcg ttc ctg cgc ggc gag ctt ggt aac cct cca ggt ggc tgg      2784
Val Ile Ala Phe Leu Arg Gly Glu Leu Gly Asn Pro Pro Gly Gly Trp
        915                 920                 925

cca gag cca ctg cgc acc cgc gca ctg gaa ggc cgc tcc gaa ggc aag      2832
Pro Glu Pro Leu Arg Thr Arg Ala Leu Glu Gly Arg Ser Glu Gly Lys
    930                 935                 940

gca cct ctg acg gaa gtt cct gag gaa gag cag gcg cac ctc gac gct      2880
Ala Pro Leu Thr Glu Val Pro Glu Glu Glu Gln Ala His Leu Asp Ala
945                 950                 955                 960

gat gat tcc aag gaa cgt cgc aat agc ctc aac cgc ctg ctg ttc ccg      2928
Asp Asp Ser Lys Glu Arg Arg Asn Ser Leu Asn Arg Leu Leu Phe Pro
                965                 970                 975

aag cca acc gaa gag ttc ctc gag cac cgt cgc cgc ttc ggc aac acc      2976
Lys Pro Thr Glu Glu Phe Leu Glu His Arg Arg Arg Phe Gly Asn Thr
            980                 985                 990

tct gcg ctg gat gat cgt gaa ttc  ttc tac ggc ctg gtc  gaa ggc cgc    3024
Ser Ala Leu Asp Asp Arg Glu Phe  Phe Tyr Gly Leu Val  Glu Gly Arg
        995                 1000                1005

gag act  ttg atc cgc ctg cca  gat gtg cgc acc cca  ctg ctt gtt       3069
Glu Thr  Leu Ile Arg Leu Pro  Asp Val Arg Thr Pro  Leu Leu Val
    1010                1015                1020

cgc ctg  gat gcg atc tct gag  cca gac gat aag ggt  atg cgc aat       3114
Arg Leu  Asp Ala Ile Ser Glu  Pro Asp Asp Lys Gly  Met Arg Asn
    1025                1030                1035
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gtt gtg  gcc aac gtc aac ggc  cag atc cgc cca atg  cgt gtg cgt      3159
Val Val  Ala Asn Val Asn Gly  Gln Ile Arg Pro Met  Arg Val Arg
    1040                 1045                 1050
gac cgc  tcc gtt gag tct gtc  acc gca acc gca gaa  aag gca gat      3204
Asp Arg  Ser Val Glu Ser Val  Thr Ala Thr Ala Glu  Lys Ala Asp
    1055                 1060                 1065
tcc tcc  aac aag ggc cat gtt  gct gca cca ttc gct  ggt gtt gtc      3249
Ser Ser  Asn Lys Gly His Val  Ala Ala Pro Phe Ala  Gly Val Val
    1070                 1075                 1080
acc gtg  act gtt gct gaa ggt  gat gag gtc aag gct  gga gat gca      3294
Thr Val  Thr Val Ala Glu Gly  Asp Glu Val Lys Ala  Gly Asp Ala
    1085                 1090                 1095
gtc gca  atc atc gag gct atg  aag atg gaa gca aca  atc act gct      3339
Val Ala  Ile Ile Glu Ala Met  Lys Met Glu Ala Thr  Ile Thr Ala
    1100                 1105                 1110
tct gtt  gac ggc aaa atc gat  cgc gtt gtg gtt cct  gct gca acg      3384
Ser Val  Asp Gly Lys Ile Asp  Arg Val Val Val Pro  Ala Ala Thr
    1115                 1120                 1125
aag gtg  gaa ggt ggc gac ttg  atc gtc gtc gtt tcc  taa              3423
Lys Val  Glu Gly Gly Asp Leu  Ile Val Val Val Ser
    1130                 1135                 1140
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<210> 65
<211> 1140
<212> PRT
<213> Corynebacterium glutamicum

<400> 65

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Val Ser Thr His Thr Ser Ser Thr Leu Pro Ala Phe Lys Lys Ile Leu
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            20                  25                  30
Glu Thr Gly Ala Ala Thr Val Ala Ile Tyr Pro Arg Glu Asp Arg Gly
        35                  40                  45
Ser Phe His Arg Ser Phe Ala Ser Glu Ala Val Arg Ile Gly Thr Glu
    50                  55                  60
Gly Ser Pro Val Lys Ala Tyr Leu Asp Ile Asp Glu Ile Ile Gly Ala
65                  70                  75                  80
Ala Lys Lys Val Lys Ala Asp Ala Ile Tyr Pro Gly Tyr Gly Phe Leu
                85                  90                  95
Ser Glu Asn Ala Gln Leu Ala Arg Glu Cys Ala Glu Asn Gly Ile Thr
            100                 105                 110
Phe Ile Gly Pro Thr Pro Glu Val Leu Asp Leu Thr Gly Asp Lys Ser
        115                 120                 125
Arg Ala Val Thr Ala Ala Lys Lys Ala Gly Leu Pro Val Leu Ala Glu
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    130                 135                 140
Ser Thr Pro Ser Lys Asn Ile Asp Glu Ile Val Lys Ser Ala Glu Gly
145                 150                 155                 160
Gln Thr Tyr Pro Ile Phe Val Lys Ala Val Ala Gly Gly Gly Gly Arg
                165                 170                 175
Gly Met Arg Phe Val Ala Ser Pro Asp Glu Leu Arg Lys Leu Ala Thr
            180                 185                 190
Glu Ala Ser Arg Glu Ala Glu Ala Ala Phe Gly Asp Gly Ala Val Tyr
        195                 200                 205
Val Glu Arg Ala Val Ile Asn Pro Gln His Ile Glu Val Gln Ile Leu
    210                 215                 220
Gly Asp His Thr Gly Glu Val Val His Leu Tyr Glu Arg Asp Cys Ser
225                 230                 235                 240
Leu Gln Arg Arg His Gln Lys Val Val Glu Ile Ala Pro Ala Gln His
                245                 250                 255
Leu Asp Pro Glu Leu Arg Asp Arg Ile Cys Ala Asp Ala Val Lys Phe
            260                 265                 270
Cys Arg Ser Ile Gly Tyr Gln Gly Ala Gly Thr Val Glu Phe Leu Val
        275                 280                 285
Asp Glu Lys Gly Asn His Val Phe Ile Glu Met Asn Pro Arg Ile Gln
    290                 295                 300
Val Glu His Thr Val Thr Glu Glu Val Thr Glu Val Asp Leu Val Lys
305                 310                 315                 320
Ala Gln Met Arg Leu Ala Ala Gly Ala Thr Leu Lys Glu Leu Gly Leu
                325                 330                 335
Thr Gln Asp Lys Ile Lys Thr His Gly Ala Ala Leu Gln Cys Arg Ile
            340                 345                 350
Thr Thr Glu Asp Pro Asn Asn Gly Phe Arg Pro Asp Thr Gly Thr Ile
        355                 360                 365
Thr Ala Tyr Arg Ser Pro Gly Gly Ala Gly Val Arg Leu Asp Gly Ala
    370                 375                 380
Ala Gln Leu Gly Gly Glu Ile Thr Ala His Phe Asp Ser Met Leu Val
385                 390                 395                 400
Lys Met Thr Cys Arg Gly Ser Asp Phe Glu Thr Ala Val Ala Arg Ala
                405                 410                 415
Gln Arg Ala Leu Ala Glu Phe Thr Val Ser Gly Val Ala Thr Asn Ile
            420                 425                 430
Gly Phe Leu Arg Ala Leu Leu Arg Glu Glu Asp Phe Thr Ser Lys Arg
        435                 440                 445
Ile Ala Thr Gly Phe Ile Ala Asp His Pro His Leu Leu Gln Ala Pro
    450                 455                 460
Pro Ala Asp Asp Glu Gln Gly Arg Ile Leu Asp Tyr Leu Ala Asp Val
465                 470                 475                 480
Thr Val Asn Lys Pro His Gly Val Arg Pro Lys Asp Val Ala Ala Pro
                485                 490                 495
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Ile Asp Lys Leu Pro Asn Ile Lys Asp Leu Pro Leu Pro Arg Gly Ser
500 505 510
Arg Asp Arg Leu Lys Gln Leu Gly Pro Ala Ala Phe Ala Arg Asp Leu
515 520 525
Arg Glu Gln Asp Ala Leu Ala Val Thr Asp Thr Thr Phe Arg Asp Ala
530 535 540
His Gln Ser Leu Leu Ala Thr Arg Val Arg Ser Phe Ala Leu Lys Pro
545 550 555 560
Ala Ala Glu Ala Val Ala Lys Leu Thr Pro Glu Leu Leu Ser Val Glu
565 570 575
Ala Trp Gly Gly Ala Thr Tyr Asp Val Ala Met Arg Phe Leu Phe Glu
580 585 590
Asp Pro Trp Asp Arg Leu Asp Glu Leu Arg Glu Ala Met Pro Asn Val
595 600 605
Asn Ile Gln Met Leu Leu Arg Gly Arg Asn Thr Val Gly Tyr Thr Pro
610 615 620
Tyr Pro Asp Ser Val Cys Arg Ala Phe Val Lys Glu Ala Ala Ser Ser
625 630 635 640
Gly Val Asp Ile Phe Arg Ile Phe Asp Ala Leu Asn Asp Val Ser Gln
645 650 655
Met Arg Pro Ala Ile Asp Ala Val Leu Glu Thr Asn Thr Ala Val Ala
660 665 670
Glu Val Ala Met Ala Tyr Ser Gly Asp Leu Ser Asp Pro Asn Glu Lys
675 680 685
Leu Tyr Thr Leu Asp Tyr Tyr Leu Lys Met Ala Glu Glu Ile Val Lys
690 695 700
Ser Gly Ala His Ile Leu Ala Ile Lys Asp Met Ala Gly Leu Leu Arg
705 710 715 720
Pro Ala Ala Val Thr Lys Leu Val Thr Ala Leu Arg Arg Glu Phe Asp
725 730 735
Leu Pro Val His Val His Thr His Asp Thr Ala Gly Gly Gln Leu Ala
740 745 750
Thr Tyr Phe Ala Ala Ala Gln Ala Gly Ala Asp Ala Val Asp Gly Ala
755 760 765
Ser Ala Pro Leu Ser Gly Thr Thr Ser Gln Pro Ser Leu Ser Ala Ile
770 775 780
Val Ala Ala Phe Ala His Thr Arg Arg Asp Thr Gly Leu Ser Leu Glu
785 790 795 800
Ala Val Ser Asp Leu Glu Pro Tyr Trp Glu Ala Val Arg Gly Leu Tyr
805 810 815
Leu Pro Phe Glu Ser Gly Thr Pro Gly Pro Thr Gly Arg Val Tyr Arg
820 825 830
His Glu Ile Pro Gly Gly Gln Leu Ser Asn Leu Arg Ala Gln Ala Thr
835 840 845
Ala Leu Gly Leu Ala Asp Arg Phe Glu Leu Ile Glu Asp Asn Tyr Ala

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    850                 855                 860
Ala Val Asn Glu Met Leu Gly Arg Pro Thr Lys Val Thr Pro Ser Ser
865                 870                 875                 880
Lys Val Val Gly Asp Leu Ala Leu His Leu Val Gly Ala Gly Val Asp
                885                 890                 895
Pro Ala Asp Phe Ala Ala Asp Pro Gln Lys Tyr Asp Ile Pro Asp Ser
            900                 905                 910
Val Ile Ala Phe Leu Arg Gly Glu Leu Gly Asn Pro Pro Gly Gly Trp
        915                 920                 925
Pro Glu Pro Leu Arg Thr Arg Ala Leu Glu Gly Arg Ser Glu Gly Lys
    930                 935                 940
Ala Pro Leu Thr Glu Val Pro Glu Glu Glu Gln Ala His Leu Asp Ala
945                 950                 955                 960
Asp Asp Ser Lys Glu Arg Arg Asn Ser Leu Asn Arg Leu Leu Phe Pro
                965                 970                 975
Lys Pro Thr Glu Glu Phe Leu Glu His Arg Arg Arg Phe Gly Asn Thr
            980                 985                 990
Ser Ala Leu Asp Asp Arg Glu Phe  Phe Tyr Gly Leu Val  Glu Gly Arg
        995                 1000                1005
Glu Thr  Leu Ile Arg Leu Pro  Asp Val Arg Thr Pro  Leu Leu Val
    1010                1015                1020
Arg Leu  Asp Ala Ile Ser Glu  Pro Asp Asp Lys Gly  Met Arg Asn
    1025                1030                1035
Val Val  Ala Asn Val Asn Gly  Gln Ile Arg Pro Met  Arg Val Arg
    1040                1045                1050
Asp Arg  Ser Val Glu Ser Val  Thr Ala Thr Ala Glu  Lys Ala Asp
    1055                1060                1065
Ser Ser  Asn Lys Gly His Val  Ala Ala Pro Phe Ala  Gly Val Val
    1070                1075                1080
Thr Val  Thr Val Ala Glu Gly  Asp Glu Val Lys Ala  Gly Asp Ala
    1085                1090                1095
Val Ala  Ile Ile Glu Ala Met  Lys Met Glu Ala Thr  Ile Thr Ala
    1100                1105                1110
Ser Val  Asp Gly Lys Ile Asp  Arg Val Val Val Pro  Ala Ala Thr
    1115                1120                1125
Lys Val  Glu Gly Gly Asp Leu  Ile Val Val Val Ser
    1130                1135                1140
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Claims

1. A method for producing an L-amino acid selected from the group consisting of L-glutamic acid, L-glutamine, L-proline, and L-arginine, comprising culturing a L-amino acid-producing bacterium which has been modified to have enhanced phosphotransacetylase activity in a medium, and collecting the L-amino acid from the medium or the bacterium.

2. The method according to claim 1, wherein the phosphotransacetylase activity is enhanced by increasing the copy

number of the gene encoding phosphotransacetylase, or by modifying an expression regulatory sequence of the gene encoding phosphotransacetylase.

3. The method according to claim 2, wherein the gene encoding phosphotransacetylase is a DNA selected from the group consisting of:

(a) a DNA comprising nucleotides 1214 to 2641 of SEQ ID NO: 34 or the nucleotide sequence of SEQ ID NO: 40; and
(b) a DNA which hybridizes with a nucleotide sequence which is complementary to the nucleotide sequence of nucleotides 1214 to 2641 of SEQ ID NO: 34, or the nucleotide sequence of SEQ ID NO: 40, wherein said DNA hybridizes under stringent conditions and encodes a protein that has phosphotransacetylase activity.

4. The method according to any one of claims 1 to 3, wherein the phosphotransacetylase activity is enhanced by disrupting a ramB gene.

5. The method according to any one of claims 1 to 4, wherein the bacterium is further modified so that an activity is enhanced of a protein selected from the group consisting of D-xylose 5-phosphate-phosphoketolase, fructose 6-phosphate phosphoketolase, and combinations thereof.

6. The method according to any one of claims 1 to 5, wherein the bacterium is further modified to enhance pyruvate carboxylase activity.

7. The method according to any one of claims 1 to 6, wherein the bacterium is further modified to have enhanced phosphoenolpyruvate carboxylase activity.

8. The method according to any one of claims 1 to 7, wherein the bacterium is selected from the group consisting of a coryneform bacterium, *Pantoea* bacterium, *Enterobacter* bacterium, and *Escherichia* bacterium.

Patentansprüche

1. Verfahren zur Herstellung einer L-Aminosäure, ausgewählt aus der Gruppe bestehend aus L-Glutaminsäure, L-Glutamin, L-Prolin und L-Arginin, umfassend das Kultivieren eines L-Aminösäureproduzierenden Bakteriums, das so modifiziert worden ist, dass es eine erhöhte Phosphotransacetylase-Aktivität aufweist, in einem Medium und Gewinnen der L-Aminosäure aus dem Medium oder dem Bakterium.

2. Verfahren gemäss Anspruch 1, worin die Phosphotransacetylase-Aktivität durch Erhöhen der Kopienzahl des Gens, das für Phosphotransacetylase codiert, oder durch Modifizieren einer expressionsregulatorischen Sequenz des Gens, das für Phosphotransacetylase codiert, erhöht ist.

3. Verfahren gemäss Anspruch 2, worin das Gen, das für Phosphotransacetylase codiert, eine DNA ist, ausgewählt aus der Gruppe bestehend aus:

(a) einer DNA, umfassend die Nukleotide 1214 bis 2641 von SEQ ID NO: 34 oder die Nukleotidsequenz von SEQ ID NO: 40; und
(b) einer DNA, die mit einer Nukleotidsequenz, die zu der Nukleotidsequenz von Nukleotiden 1214 bis 2641 von SEQ ID NO: 34 oder der Nukleotidsequenz von SEQ ID NO: 40 komplementär ist, hybridisiert, wobei die DNA unter stringenten Bedingungen hybridisiert und ein Protein codiert, das Phosphotransacetylase-Aktivität aufweist.

4. Verfahren gemäss einem der Ansprüche 1 bis 3, worin die Phosphotransacetylase-Aktivität durch Unterbrechen eines ramB-Gens erhöht ist.

5. Verfahren gemäss einem der Ansprüche 1 bis 4, worin das Bakterium des weiteren modifiziert ist, so dass die Aktivität eines Proteins, ausgewählt aus der Gruppe bestehend aus D-Xylose-5-phosphat-phosphoketolase, Fructose-6-phosphat-phosphoketolase oder Kombinationen davon, erhöht ist.

6. Verfahren gemäss einem der Ansprüche 1 bis 5, worin das Bakterium des weiteren modifiziert ist, um die Pyruvat-

carboxylase-Aktivität zu erhöhen.

7. Verfahren gemäss einem der Ansprüche 1 bis 6, worin das Bakterium weiter modifiziert ist, um eine erhöhte Phosphoenolpyruvatcarboxylase-Aktivität aufzuweisen.

8. Verfahren gemäss einem der Ansprüche 1 bis 7, worin das Bakterium ausgewählt ist aus der Gruppe bestehend aus einem coryneformen Bakterium, Pantoea-Bakterium, Enterobacter-Bakterium und Escherichia-Bakterium.

Revendications

1. Procédé pour produire un acide L-aminé choisi dans le groupe constitué de l'acide L-glutamique, la L-glutamine, la L-proline, et la L-arginine, comprenant la culture d'une bactérie produisant un acide L-aminé qui a été modifiée pour avoir une activité phosphotransacétylase améliorée dans un milieu, et la collecte de l'acide L-aminé depuis le milieu ou la bactérie.

2. Procédé selon la revendication 1, dans lequel l'activité phosphotransacétylase est améliorée en augmentant le nombre de copies du gène codant pour la phosphotransacétylase, ou en modifiant une séquence régulatrice d'expression du gène codant pour la phosphotransacétylase.

3. Procédé selon la revendication 2, dans lequel le gène codant pour la phosphotransacétylase est un ADN choisi dans le groupe constitué de :

   (a) un ADN comprenant les nucléotides 1214 à 2641 de SEQ ID NO: 34 ou la séquence nucléotidique de SEQ ID NO: 40 ; et
   (b) un ADN qui s'hybride avec une séquence nucléotidique qui est complémentaire de la séquence nucléotidique des nucléotides 1214 à 2641 de SEQ ID NO: 34, ou la séquence nucléotidique de SEQ ID NO: 40, dans lequel ledit ADN s'hybride dans des conditions stringentes et code pour une protéine qui possède une activité phosphotransacétylase.

4. Procédé selon l'une quelconque des revendications 1 à 3, dans lequel l'activité phosphotransacétylase est améliorée en interrompant un gène ramB.

5. Procédé selon l'une quelconque des revendications 1 à 4, dans lequel la bactérie est en outre modifiée de sorte qu'une activité est améliorée d'une protéine choisie dans le groupe constitué de la D-xylose 5-phosphate-phosphocétolase, la fructose 6-phosphate-phosphocétolase, et les combinaisons de celles-ci.

6. Procédé selon l'une quelconque des revendications 1 à 5, dans lequel la bactérie est en outre modifiée pour améliorer l'activité pyruvate carboxylase.

7. Procédé selon l'une quelconque des revendications 1 à 6, dans lequel la bactérie est en outre modifiée pour avoir une activité phosphoénolpyruvate carboxylase améliorée.

8. Procédé selon l'une quelconque des revendications 1 à 7, dans lequel la bactérie est choisie dans le groupe constitué d'une bactérie corynéforme, une bactérie *Pantoea,* une bactérie *Enterobacter,* et une bactérie *Escherichia.*

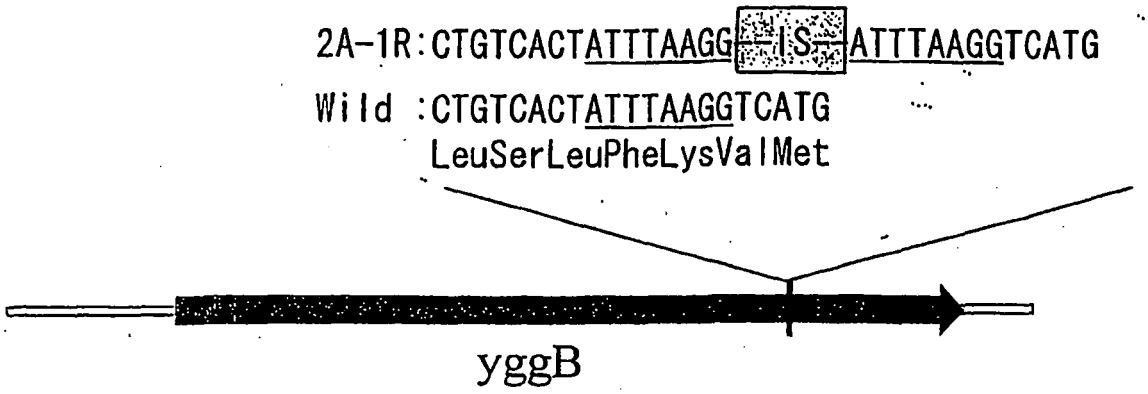

2A-1R:CTGTCACTATTTAAGGISATTTAAGGTCATG
Wild :CTGTCACTATTTAAGGTCATG
LeuSerLeuPheLysValMet
yggB

Fig. 1

REFERENCES CITED IN THE DESCRIPTION

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